Journal of Aquaculture Feed Science and Nutrition 6 (1): 23-31, 2014
ISSN: 2070-1667

Effect of Live Feed *Mesocyclops aspericornis* Survival, Growth, Biochemical Constituents and Energy Utilization of the Freshwater Fish *Catla catla*

S. Munirasu, V. Uthayakumar, V. Ramasubramanian and A. Kiruba
Unit of Aquatic Biotechnology and Live feed Culture Lab, Department of Zoology,
Bharathiar University, 641046 Coimbatore, Tamil Nadu, India

Abstract: This experiment was carried out to evaluate the effects of the *Mesocyclops aspericornis* enriched with different micro algae, such as *Spirulina platensis*, *Chlorella vulgaris* and *Spirogyra maximus* on survival, growth, biochemical constituents and energy utilization of freshwater fish *Catla catla*. Experimental diets were the same in all, except for the inclusion in the experimental diets. The experimental fishes were fed with Spirulina enriched Cyclops (E2), Chlorella enriched Cyclops (E3), Spirogyra enriched Cyclops (E4) and unenriched Cyclops (E1) as a control and triplicates were maintained for each treatments. These diets were fed to fries of *C. catla* for a period of 60 days. The growth parameters, such as survival, weight gain, specific growth rate, feed conversion efficiency and protein efficiency rate were significantly ($p<0.05$) higher in (E2), Spirulina enriched Cyclops incorporated diet fed fries followed by other experimental groups than the control. Similarly, the biochemical constituents of the protein, amino acid, carbohydrate, lipid and ash content were significantly ($p<0.05$) higher in (E2), Spirulina enriched Cyclops incorporated diet fed fries fish. The energy utilization parameters, such as feeding rate, absorption rate, conversion rate and metabolic rate were significantly ($p<0.05$) higher in (E2) Spirulina enriched Cyclops incorporated diet fed fries fish. From the results of this experiment, researchers conclude that Spirulina enriched Cyclops was the best algal food for the fries of *C. catla* larval culture. Hence, researchers conclude that *S. platensis* enriched *M. aspericornis* a better live food for the larval rearing of commercial fish *C. catla*.

Key words: *Spirulina platensis*, *Chlorella vulgaris*, *Catla catla*, *Spirogyra maxima*, nutritional values *Mesocyclops aspericornis*

INTRODUCTION

Fingerling production is one of the hallenges faced by those interested in promoting production of emerging marine and fresh water species. In spite of huge efforts to use artificial feeds, the culture of fish larvae during the primary nursing phase still depends heavily on natural food. Live feeds include Cyclops, Artemia and other tiny organisms are often the first foods in the aquaculture food chain (Sorgeloos, 1980; Leger *et al*., 1986).

Copepods have been recognized as the most suitable feed for early stages of fish larvae because of their nutritional status high essential fatty acids compared with other live feeds, such as rotifers and artemia (Nanton and Castell, 1998; Evjemo *et al*., 2003; Stottrup and McEvoy, 2003). Copepods have been mass-cultured, as early as 1970 and 1980s in Japan and have been used as feed for Pacific cod larvae in rearing trials in fisheries stations (Hagiwara *et al*., 2001). Different algae species possess diverse intrinsic biochemical features, making them suitable for some grazers but not for others. Diatoms, for instance are often considered as high-quality food for copepods (Jonasdottir and Kiorboe, 1996). Nevertheless, positive nutritional effects of copepods on marine finfish have been reported, such as increased growth and survival (Doi *et al*., 1997; Nanton and Castell, 1998; Stottrup and Jensen, 1990; Copeman *et al*., 2002; Skalli and Robin, 2004), improved pigmentation (Bell *et al*., 2003), retinal morphology (Shields *et al*., 1999), broodstock reproductive performance and egg and larval quality (Mazorra *et al*., 2003).

Spirulina is considered a rich source of protein, vitamins, minerals, essential amino acids and fatty acids, Gamma-Linolenic Acid (GLA) and antioxidant pigments, such as carotenoids (Belay *et al*., 1996). In addition, it is effective as an immune modulator (Takeuchi *et al*., 2002).

Previous studies have also found *Rhodomonas* sp. to be an excellent food source for copepod species (Koski *et al*., 1998; Rey *et al*., 2001; Knuckey *et al*., 2005).

Corresponding Author: V. Ramasubramanian, Unit of Aquatic Biotechnology and Live feed Culture Lab, Department of Zoology, Bharathiar University, 641046 Coimbatore, Tamil Nadu, India

One reason for the high nutritional quality of *R. salina* may be its high content of PUFAs. It is believed that PUFAs are one of the important aspects of food quality (Romdhane *et al*., 1995; Gonzalez-Felix *et al*., 2003). The HUFAs have additionally been shown to have an even greater essential FA value than the PUFAs for crustacean survival, growth, metamorphosis rate and stress resistance (Romdhane *et al*., 1995; Gonzalez-Felix *et al*., 2003). In *R. salina*, a high HUFA content (39%) was likely to be an important reason for the growth success of copepods feeding on this alga. The advantages of algae as a food is enormous as algal feeds are easy to culture and it is an excellent feed for the growth of zooplanktons (Bogdan and Gilbert, 1987). Algae are alternative plant feedstuffs that are increasingly being used in aqua feeds because of their nutritional quality, low cost and availability (Mustafa and Nakagawa, 1995). Although, Artemia enrichment has greatly improv ed commercial aquaculture, the most advanced rotifer and Artemia bioencapsulation techniques have matched the results with copepods and their composition is now taken, as the standard for improving enrichment techniques for rotifers and Artemia (Kraul *et al*., 1988, 1992; Sorgeloos and Leger, 1992). Algae are the main sources of Highly Unsaturated Fatty Acids (HUFA) for zooplankton (Witt *et al*., 1984; Kanazawa *et al*., 1985; Koven *et al*., 1992; Sargent *et al*., 1997). Enrichment or boosting of the aquatic feeds into the organisms has been incorporated in to the larval rearing protocols for many fish species (Sorgeloos *et al*., 1991). Cyclops which is incapable of synthesizing highly polyunsaturated fatty acid and essential amino acids, it needs to be enriched by micro algal enrichment which contain high level of amino acid in particular has high biological value during larval development (Harish and Gajaria, 1995). *Catla catla* is the fastest growing species (Ravi and Devaraj, 1991).

The aim of the present investigation is to evaluate the effect of *M. aspericornis* enriched with different algae feeds phytoplankton's (*S. platensis*, *C. vulgaris* and *S. maxima*) on survival and growth of *C. catla* fries.

MATERIALS AND METHODS

The fresh water *C. catla* fries were collected from Bhavani Sagar Government Fish Development Corporation, Erode District, Tamil Nadu. They were transported safely and brought to the laboratory in well-oxygenated plastic bags. They were stocked in large cement tank (6×4×3) and acclimatized to the laboratory condition for 2 weeks before the commencement of experiments. During acclimatization, *C. catla* were fed with commercial feed. Water was routinely changed every day in order to maintain a healthy environment for the fishes apart from providing artificial aeration. The experimental water presented the following physicochemical parameters: pH, 7.10±0.50; total dissolved solids, 0.98±0.10 g L^{-1}, dissolved oxygen, 7.30±0.40 mg L^{-1}, BOD, 40.00±1.60 mg L^{-1}, COD, 120.00±9.00 mg L^{-1} and ammonia, 0.068±0.008 mg L^{-1}.

Experimental setup: The fresh water fish fries *C. catla* with initial length of 1.6±0.15 cm and initial weights 0.215±0.11 g were used. The experimental period was restricted to 60 days. Each experimental trough contained 40 L of water capacity. The experimental groups were fed with different algae, control fishes were fed with unenriched Cyclops (E1) and experimental fishes were fed with *Spirulina platensis* enriched Cyclops (E2), fishes were fed with *Chlorella vulgaris* enriched Cyclops (E3), fishes were fed with *Spirogyra maximus* enriched Cyclops (E4). Before initializing the experiment, the initial length and weight of the animals were measured, similarly at the end of this experiment (on 60th day) the final length and weight were measured.

Phytoplankton culture: In the present study important algae, such as *S. platensis, C. vulgaris* and *S. maxima* were cultured to enrich the Zooplankton (*M. aspericornis*).

Collection of phytoplankton: The experimental micro algae, *S. platensis* and *C. vulgaris* pure culture were obtained from Antenna Green Trust, Antheneri village, Kadachanenthal, Madurai, Tamil Nadu. The Spirogyra cultures were collected from IRTC, Palakkad, Kerala. The pure cultures were transferred in oxygen filled polyethylene covers with proper aeration.

Culture of *Spirulina platensis*: The pure Spirulina cultures were poured into the 24 L plastic tubs and were placed in sun light. Several physico-chemical parameters were maintained in culture medium for the proper culture of Spirulina such as pH, temperature and dissolved solids. The parameters were checked at every 2 days interval. Proper aeration was provided through aerators for proper mixing of chemicals and proper growth of Spirulina.

Culture of *Chlorella vulgaris*: The fresh water green *C. vulgaris* were cultured in aquarium tanks. The salt and biochemical were dissolved in ground water. After that, cow dung was mixed with water and filtered with the nylon cloth. The filtrate was put in the culture and mixed well during the culture period. The pH was maintained at 8 and no aeration was provided. The pure *C. vulgaris* culture were poured into the 24 L plastic tubs and placed in sun light. The culture medium was stirred twice daily for proper distribution of chemicals and good aeration, such as pH, temperature and dissolved solids. To prevent

sedimentation and a homogenous exposure of algal cells to light, reduce the nutrient and temperature gradient along depth of culture.

Culture of *Spirogyra maxima*: The algal samples were collected kavery river just below the water surface in 500 mL plastic containers with screw caps. The Spirogyra was collected and washed thoroughly to remove the adhering dirt. Fresh water mixed biochemical nutrient, added mother *Spirogyra maxima* culture.

Calculation of growth rate in phytoplankton: After the 20 days, *S. platensis*, *C. vulgaris* and *S. maxima* can be viewed as a thin layer on the surface of the medium. At this stage, the Spirulina cell count may be calculated by the following equation:

$$r = \frac{\ln(N_t) - \ln(N_o)}{t}$$

Where:

N_t = Final density of phytoplankton
N_o = Initial density of phytoplankton
t = Time interval between the initial and final density estimated
In = Individual animals
r = Growth rate

Procedures were repeated in several tubs and required quantities of phytoplankton were cultivated.

Harvesting of phytoplankton: Productivities rarely exceed 30-200 g day^{-1} and cell densities of 2 g L^{-1}. The Spirulina and Chlorella were harvested by filtration through meshes having size about 10 μm. After harvesting, the algal biomass was dehydrated by sun during. The harvested Spirulina and Chlorella powders were stored in clean and air tight containers. During weight measurements of algae powders were calculated in gram per liter by during the biomass in an oven at 105°C for 2 h. This Spirulina and Chlorella powders were used to enrich the Cyclops.

Harvesting and drying of Spirogyra: The cultured Spirogyra were filtered with the help of net of mesh size 30 μm and washed thoroughly with the clean water and dried in a hot oven at 60°C. After drying Spirogyra were powdered into fine particles, properly weighed and then stored in clean containers for further use.

Zooplankton culture: The live feed of present investigation Cyclops collected from Muthanna Lake, Coimbatore, Tamil Nadu, India. During collection period the dip net was swept through the surface water near the shore. Collected mixed zooplanktons were stored in a container. The sample was diluted (5 times) by adding water. Using the plankton net Cyclops was isolated from other zooplanktons. Transferred to a cement tank which was prefilled with soil (5 cm depth), poultry manure (0.4 kg) lime powder (1 kg) and water of 15 cm height for further culture. The density of Cyclops were calculated as follows:

$$r = \frac{\ln(N_t) - \ln(N_o)}{t}$$

Where:

N_t = Final density of the Cyclops
N_o = Initial density of Cyclops
t = Time interval between the initial and final density estimation
In = Individual animals

Enrichment: Cyclops was enriched with *S. platensis*, *C. vulgaris* and *S. maxima* to feed the commercially important fish fries of *C. catla*, cultured in laboratory. The 48 h adult nauplii of Cyclops were fed with each type of food at same concentration 0.5 mg/mL/day. The powdered feeds are taken at 0.5 mg concentration, mixed with distilled water and stirred for 2-3 min vigorously. Cyclops (50 mL^{-1}) was introduced into 500 mL culture flasks containing freshwater and mild aeration was provided. After 6 h of enrichment Cyclops adult nauplii were fed to experimental fishes *C. catla* twice a day. Daily observations were done. After 60 days, the final length, weight and percentage of survival were determined.

Nutritional analysis: Tissues taken from the fries of *C. catla* were analyses for biochemical parameters. Following the basic procedures of Lowry *et al.* (1951) for protein, Folch *et al.* (1957) for lipids and Roe (1955) for carbohydrates. Fatty acid analysis was done using gas chromatography described by Nichols *et al.* (1986).

Growth analysis: The growth parameters were calculated by using the following equation according to (Felix and Sudharsan 2004; Venkat *et al.*, 2004):

$$\text{Weight gain}(g) = \text{Final weight}(g) - \text{Initial weight}(g)$$

$$\text{Specific Growth Rate}(SGR) = \frac{\text{Final weight}(g) - \text{Initial weight}(g)}{\text{Initial weight}(g)} \times 100$$

$$\text{Food Conversion Ratio}(FCR) = \frac{\text{Total feed given}(g)}{\text{Total weight gain fish}(g)} \times 100$$

$$\text{Food Conversion Efficiency}(\%)(\text{FCE}) = \frac{\text{Biomass}(g)}{\text{Total feed intake}(g)} \times 100$$

$$\text{Feeding Rate}(\text{FR}) = \frac{\text{Mean food consumption}(\text{k.cal/day})}{\text{Initial live weight of the fishes}(g)}$$

$$\text{Survival}(\%) = \frac{\text{No. of survived at the end of the experiment}}{\text{No. of fishes stocked at the start of the experiment}} \times 100$$

$$\text{Metabolic Rate}(\text{MR}) = \text{Absorption rate}(\text{k.cal/g/day}) - \text{Conversion rate}(\text{k.cal/g/day}) + NH_3 \text{ excretion rate}(\text{k.cal/g/day})$$

Statistical analysis: Statistical analysis was performed using Analysis of Variance (one-way ANOVA) and Student's t-test, to determine differences between experimental levels. Levels of significance are expressed as $p<0.05$. All analyses were performed using the Statistical Analysis System (SAS) program.

RESULTS AND DISCUSSION

Growth parameters: The combined different algal enrichment of *M. aspericornis* diets fed fries of *C. catla* fish morphometric, survival and growth performance data's were showed in Table 1. The initial average body length and weight of was 1.6±0.15 cm and 0.215±0.11 g. After the feeding experiment, the morphometric data (4.71±0.22 and 3.43±0.33 cm), survival and growth performance (WG, SGR, FCE and PER) were significantly ($p<0.05$) higher in (E2) Spirulina enriched Cyclops incorporated diets, followed by the fries fed with E3, E4 and control E1. The FCR was found to be lower in fries fed with E2 supplemented diet ($p<0.05$) followed by the fries fed with E3, E4 and the control E1 (Fig. 1).

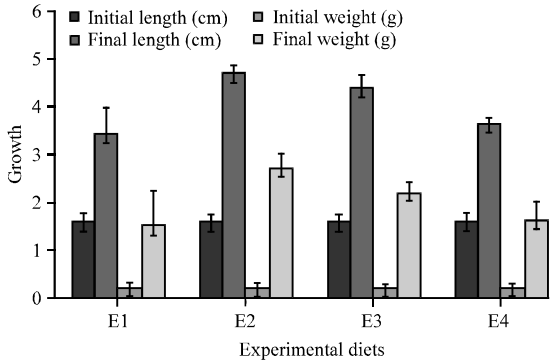

Fig. 1: The growth performance of *C. catla* fed with different algal enrichment of *M. aspericornis* (Mean±SD)

Biochemical-analysis of experimental diets and tissues: Chemical compositions of the experimental tissues were reported in Table 1. After the feeding experiment, the protein, lipid and carbohydrate were significantly ($p<0.05$) higher in E2 Spirulina enriched Cyclops incorporated diets, followed by the fries fed with E3, E4 and control E1. Variation in the values of crude protein content of feed was (43.3±2.86 to 52.91±7.06%), crude lipid content (22.17±4.57 to 31.94±7.79%) crude carbohydrate content (32.34±4.65 to 38.68±3.82%), ash content (15.15±2.71 to 19.24±4.17) and moisture content (69.32±1.24 to 75.07±2.17) recorded, respectively (Fig. 2).

Energy utilization performance: The energy utilization performance of different algal enrichment of *M. aspericornis* diets fed *C. catla* fries fish was also given in Table 1. The feeding rate, absorption rate, conversion rate, NH_3 execratory rate and metabolic rate were found to be maximum in fed with E2, Spirulina enriched Cyclops incorporated diet fed fingerlings fish, followed by the fed with E3, E4 and compared with control E1. The statistical analysis made on the energy utilization performance between control and experimental diets revealed that the variation between them was significant ($p<0.05$) (Fig. 3).

Fatty acid profile: Fatty acids profile of *C. catla* fed with different phytoplankton enriched *M. aspericornis* (expressed in mole %) were tabulated in Table 2. In the *S. platensis* enriched *M. aspericornis* the total Saturated Fatty Acid content was (SFA) 47.74%, total MUFA was 24.59%, total PUFA and HUFA was 42.24 and 39.0%. The *C. vulgaris* enriched *M. aspericornis* had total SFA 45.04%, total MUFA as 16.15%, total PUFA and HUFA as 38.05 and 28.9%. In the *S. maxima* enriched *M. aspericornis*, the total SFA content was 42.34%, total MUFA was 15.56%, total PUFA and HUFA was 37.31%, 15.1% and unenriched *M. aspericornis* had the total SFA as 34.76%, total MUFA as 14.6%, total PUFA and HUFA as 34.83 and 16.6%, respectively.

Table 1: The morphometric data, growth performance, biochemical constituents and energy utilization of *C. catla* fed with different algal enrichment of *M. aspericornis* (Mean±SD)

		Experimental diets			
		Control	Different algae enrichment of *M. aspericornis*		
Aspects	Parameters	E1	E2	E3	E4
Morphometry	Initial length (cm)	1.6±0.15	1.6±0.15	1.6±0.15	1.6±0.15
	Final length (cm)	3.43±0.33^{b}	4.71±0.22^{a}	4.41±0.47^{a}	3.65±0.33^{b}
	Initial weight (cm)	0.215±0.11	0.215±0.11	0.215±0.11	0.215±0.11
	Final weight (cm)	1.52±0.21	2.73±0.21a	2.23±0.17^{b}	1.64±0.28
Nutritional indices	Survival rate (%)	92.62±1.48^{c}	98.67±0.79^{a}	95.36±2.21^{b}	93.13±1.65^{bc}
	Weight gain (g)	1.14±0.21^{b}	2.66±0.63^{a}	2.33±0.73^{a}	1.39±0.34^{b}
	SGR (%)	2.07±0.34^{b}	3.29±0.61^{a}	2.76±0.75^{ab}	2.55±0.74^{ab}
	FCR (g)	2.45±0.49^{b}	3.48±0.49^{a}	2.67±0.40^{ab}	2.61±1.01^{b}
	FCE (%)	1.19±0.66^{b}	2.53±0.60^{a}	1.50±0.28^{b}	1.48±0.29^{b}
	PEC (g)	0.98±0.34^{b}	2.33±0.54^{a}	1.22±0.42^{b}	1.10±0.27^{b}
Biochemical constituents	Protein (%)	43.3±2.86^{b}	52.91±7.06^{a}	47.38±2.12^{ab}	46.46±5.43^{ab}
	Lipid (%)	22.17±4.57^{b}	31.94±7.79^{a}	27.20±2.62^{ab}	24.44±3.51^{b}
	Carbohydrate (%)	32.34±4.65^{b}	38.68±3.82^{a}	36.17±4.60^{ab}	34.44±3.76^{ab}
	Amino acid (%)	28.33±3.46^{b}	38.17±5.10^{a}	36.16±6.28^{a}	35.64±3.82^{a}
	Ash (%)	15.15±2.71^{a}	19.24±4.17^{a}	15.90±1.34^{a}	15.10±2.61^{a}
	Moisture (%)	69.32±1.24^{b}	75.07±2.17^{a}	71.05±2.17^{b}	70.26±2.01^{b}
Energy utilization (k.cal/g/day)	Feeding rate	0.374±0.183^{b}	0.641±0.252^{a}	0.516±0.150^{ab}	0.502±0.141^{ab}
	Conversion rate	0.162±0.717^{b}	0.517±0.210^{a}	0.326±0.175^{ab}	0.325±0.164^{ab}
	NH_3 excretion	0.032±0.006^{c}	0.061±0.006^{a}	0.048±0.009^{b}	0.035±0.008^{c}
	Metabolic rate	0.168±0.101^{b}	0.388±0.006^{a}	0.287±0.063^{a}	0.105±0.067^{b}

Each value is a mean±SD of three replicate. Within a row, values with different superscripts are significantly different ($p<0.05$)

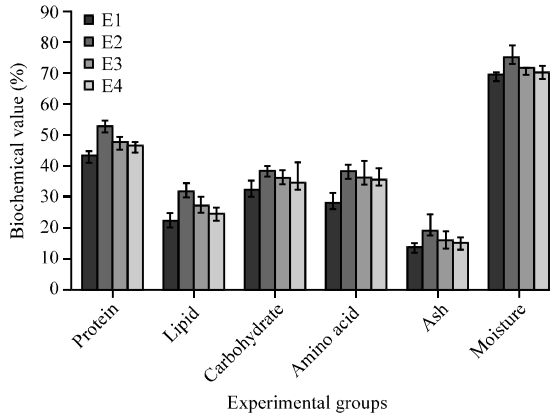

Fig. 2: The biochemical constituents of *C. catla* fed with different algal enrichment of *M. aspericornis*

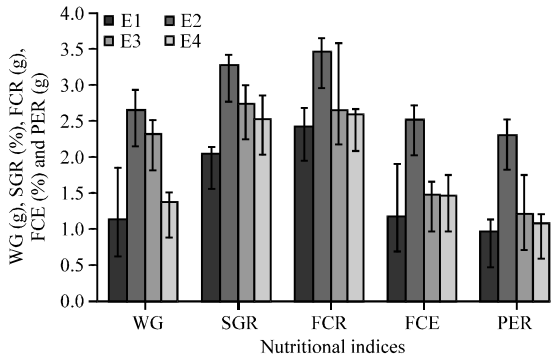

Fig. 3: The nutritional indices of *C. catla* fed with different algal enrichment of *M. aspericornis* (Mean±SD)

In the present study recommends that algae enriched copepod (*M. aspericornis*) can be exclusively used as an important alternate or supplementary feed for commercial seed production of *C. catla*. Larvae of nearly all marine and of many freshwater fish species require live feed organisms as first food for many fish species live food still gives better results in terms of growth and survival than artificial diets (Dabrowski, 1984). The best survival and growth of some fish larvae were obtained with copepods reared with N yeast (Fukusho *et al*., 1980), possibly by their higher nutritional value due to their high eicosapentaenoic acid (EPA; 20:5 (n-3)), DHA and total n-3 HUFA content (Watanabe, 1993; Fukusho *et al*., 1980).

In the present study, variations in nutritional composition among planktons were observed. The studies indicated that the sufficient densities of *S. platensis* are important for the normal growth and development of larval *C. catla* during the early development (Lu and Takeuchi, 2004). The *S. platensis* contains high protein compared to *C. vulgaris* and *S. maxima*. The micro algae, Spirulina seemed to be a good protein source (60-70%) and is comparable with the milk proteins (Shelef and Soeder, 1980). Spirulina is considered a rich source of protein, vitamins, minerals, essential amino acids and fatty acids, Gamma-Linolenic Acid (GLA) and antioxidant pigments, such as carotenoids (Belay *et al*., 1996; Phang *et al*., 2000). The

Table 2: Fatty acids profile of *C. catla* fed with different algal enriched *M. aspericornis* (expressed in mole (%))

	Experimental diets			
	Control	Different algae enrichment of *M aspericornis*		
Fatty acids	E1	E2	E3	E4
14:0	8.03	12.40	12.63	6.45
15:0	4.02	3.09	3.45	1.35
16:0	21.03	29.30	25.03	27.70
17:0	1.34	0.50	1.53	1.04
18:0	0.34	2.45	2.40	5.80
SFA	34.76	47.74	45.04	42.34
16:1	0.89	3.37	0.93	0.63
17:1	0.66	0.72	0.82	0.45
18:1 n7	4.18	4.40	5.20	5.20
18:1 n9	8.87	16.10	9.20	9.28
MUFA	14.60	24.59	16.15	15.56
18:2 n6	12.04	16.47	17.45	12.93
20:2 n6	5.04	6.45	4.28	3.34
20:3 n3	1.46	2.03	0.72	0.59
20:4 n6	7.34	7.45	4.82	5.54
20:5 n3	5.35	2.54	5.72	6.33
22:5	2.04	2.48	3.03	3.24
22:6 n3	1.56	4.81	2.03	5.34
PUFA	34.83	42.23	38.05	37.31
HUFA	16.60	39.00	28.90	15.10

biochemical composition of *S. platensis* revealed that they have high amount of protein between 55-70% depending on the source *S. platensis* is rich in high quality protein, vitamins, minerals and many biologically active substances (Becker, 1994). *S. platensis* can be used in the diets of domestic animals (Venkataraman, 1972).

After 60 days of experiment, the biochemical composition of fish enriched with *S. platensis* enriched Cyclops was found to contain maximum level of protein. Despite on the high nutritive value of algae, little information has been published on their use as a protein source for fishes (Appler, 1985; Nakagawa *et al*., 1987; Cho and Kaushik, 1990). These results indicated that *S. platensis* is best food for enrichment of *M. aspericornis* suitable for the growth and survival of commercial fish *C. catla* (Nandeesha *et al*., 1993). There is great difference found between the feeds enriched fed fish and control. Protein is the most important and expensive component of the aquaculture diets. Protein is required in the diet to provide indispensable amino acids and nitrogen for synthesis of dispensable amino acids (Balazs *et al*., 1973; Colvin and Brand, 1977). They supply the major portion of energy required by living cells. If a large percentage of the metabolic energy requirements of the animal can be met from the carbohydrate, it have the potential for delivering a low cost source of energy that could spare protein for growth (Simon, 2009). Lipids are substances found in both plants and animals (Harrison, 1990). Lipids fall into two basic categories (glycerol-based and nonglycerol-based). The lipids are important sources of metabolic energy Adenosine Triphosphate (ATP) and are the most energy-rich of all classes of nutrients (Pillay and Nair, 1973; Galois, 1984). Among the 17 protein in body tissues, 10 amino acids are essential and must be supplied through the diet since animals including fish cannot synthesis them (Watanabe, 1993; Coloso and Cruz, 1980; Kanazawa and Teshima, 1981). A large proportion of the amino acid consumed by animals is catabolized for energy. Amino acids play important and versatile roles in fish nutrition and metabolism (Li *et al*., 2009). Since, high protein diets are needed for good growth of most aquatic animals, estimation of minimum requirements of Essential Amino Acids (EAA) is indispensable to formulate cost effective diets. The quantitative EAA requirements of fish and crustaceans are often determined by feeding experiments with diets containing graded levels of the particular amino acid to be examined (Tang and Hwang, 1966). Both the absolute amounts of individual fatty acids and their relative proportion are important in the nutrition of fish larvae (Sargent *et al*., 1997). In particular, the DHA/EPA ratio may affect larval growth and survival, possibly because high amounts of EPA in relation to DHA may create an imbalance in the structural composition of the phospholipids that are essential components of biological membranes (Rainuzzo *et al*., 1994). In the present study, DHA/EPA ratios ranged from 4.7:1 for *A. sinjiensis* to 2.2:1 for *P. crassirostris*. All 3 species, therefore met or exceeded the recommended DHA/EPA ratio of about 2:1 formarine finfish larval feeds (Sargent *et al*., 1997). The improvements in larval growth, survival and rates of normal are generally attributed to levels of DHA, EPA and or Arachidonic Acid (ARA) in the diet (Tocher *et al*., 1994; Reitan *et al*., 1997).

CONCLUSION

The freshwater cyclops, *M. aspericornis* is an ideal live food for the first few days' culture of most fish larvae because of its numerous characteristics, small size, slow morbidity and easy catchability by the larvae. It is also, imported to enrich this Cyclops for all round best performance in the larva. The culture of the freshwater Cyclops, *M. aspericornis* can be maintained continuously in a feed back culture system. Many studies have been written on the usefulness of Cyclops for the raising of fish fry and how one can expect to raise a high percentage of the spawned fishes and end up with quality specimens from feeding Cyclops. Cyclops are also used as a conditioning food to induce adult fish to spawn. Freshwater larviculturist will avail themselves with the findings reported in this review in order to improve larval

performance, increase yield and facilitate breeding of new fish species. This will ensure an overall satisfactory performance in hatchery operations.

REFERENCES

Appler, N.H., 1985. Evaluation of *Hydrodictyun reticulaturn* as a protein source for *Oreochromis* (*Tiiapia*) *nilotica* and *Tilapia zillii*. J. Fish Biol., 27: 327-333.

Balazs, G.H., E. Ross and C.C. Brooks, 1973. Preliminary studies on the preparation and feeding of crustacean diets. Aquacult., 2: 369-377.

Becker, E.W., 1994. Microalgae. In: Nutrition, Becker, E.W. (Ed.) Cambridge University Press, Cambridge, pp: 196-249.

Belay, A., T. Kato and Y. Ota, 1996. Spirulina (Arthrospira): Potential application as an animal feed supplement. J. Applied Phycol., 8: 303-311.

Bell, J.G., L.A. McEvoy, A. Estevez, R.J. Shields and J.R. Sargent, 2003. Optimising lipid nutrition in first-feeding flatfish larvae. Aquaculture, 227: 211-220.

Bogdan, K.G. and J.J. Gilbert, 1987. Quantitative comparison of food niches in some freshwater zooplankton. Oecologia, 72: 331-340.

Cho, C.Y. and S.J. Kaushik, 1990. Nutritional energetics in fish: Energy and protein utilization in rainbow trout (*Salmo gairdneri*). World Rev. Nutr. Diet., 61: 132-172.

Coloso, R.M. and L.J. Cruz, 1980. Preliminary studies in some aspects of amino acid biosynthesis in juveniles of *Penaeus monodon* Fabricus I- Incorporation of 14C from (U-14C) acetate in amino acids of perceptible proteins. Bull. Phil. Biochem. Soc., 3: 12-22.

Colvin, L.B. and C.W. Brand, 1977. The protein requirement of penaeid shrimp at various life-cycle stages in controlled environment systems. Proc. Annu. Meet. World Mariculture Soc., 8: 821-840.

Copeman, L.A., C.C. Parrish, J.A. Brown and M. Harel, 2002. Effects of docosahexaenoic, eicosapentaenoic and arachidonic acids on the early growth, survival, lipid composition and pigmentation of yellowtail flounder (*Limanda ferruginea*): A live food enrichment experiment. Aquaculture, 210: 285-304.

Dabrowski, K., 1984. Influence of initial weight during the change from live to compound feed on the survival and growth of four cyprinids. Aquaculture, 40: 27-40.

Doi, M., J.D. Toledo, M.S.N. Golez, M. de los Santos and A. Ohno, 1997. Preliminary investigation of feeding performance of larvae of early red-spotted grouper, *Epinephelus coioides*, reared with mixed zooplankton. Hydrobiologia, 358: 259-263.

Evjemo, J.O., K.I. Reitan and Y. Olsen, 2003. Copepods as live food organisms in the rearing of Atlantic halibut larvae (*Hippoglossus hippoglossus* L.) with special emphasis on nutritional value. Aquacult., 227: 191-210.

Felix, N. and M. Sudharsan, 2004. Effect of glycine betaine, a feed attractant affecting growth and feed conversion of juvenile freshwater prawn *Macrobrachium rosenbergii*. Aquacult. Nutr., 10: 193-197.

Folch, J., M. Lees and G.H.S. Stanley, 1957. A simple method for the isolation and purification of total lipides from animal tissues. J. Biol. Chem., 226: 497-509.

Fukusho, K., T. Arakawa and T. Watanabe, 1980. Food value of a copepod, *Tigriopus japonicas*, cultured with Nyeast for larvae and juveniles of mud dab *Limanda yokohamae*. Bull. Jpn. Soc. Sci. Fish., 46: 499-503.

Galois, R.G., 1984. Variations de la composition lipidique tissulaire au cours de la vitellogenese chez la crevette *Penaeus indicus* Milne Edwards. J. Exp. Mar. Biol. Ecol., 84: 155-166.

Gonzalez-Felix, M.L., A.L. Lawrence, D.M. Gatlin and M. Perez-Velazquez, 2003. Nutritional evaluation of fatty acids for the open thelycum shrimp, *Litopenaeus vannamei*: I. Effect of dietary linoleic and linolenic acids at different concentrations and ratios on juvenile shrimp growth, survival and fatty acid composition. Aquacult. Nutr., 9: 105-113.

Hagiwara, A., W.G., Gallardo, M. Assavaaree, T. Kotani and A.B. de Araujo, 2001. Live food production in Japan: Recent progress and future aspects. Aquaculture, 200: 111-127.

Harish, K.M. and S.C. Gajaria, 1995. Growth of Indian major carps fed on *Spirogyra* sp. diets. Proceedings of the National Symposium on Algal Potential and Its Exploitation, December 27-29, 1995, Madurai, India, pp: 46-46.

Harrison, K., 1990. The role of nutrition in maturation, reproduction and embryonic development of decapod crustaceans. A review. J. Shellfish Res., 9: 1-28.

Jonasdottir, S.H. and T. Kiorboe, 1996. Copepod recruitment and food composition: Do diatoms affect hatching success? Mar. Biol., 125: 743-750.

Kanazawa, A. and S. Teshima, 1981. Essential amino acids of the prawn. Bull. Jpn. Soc. Sci. Fish., 17: 1375-1379.

Kanazawa, A., S.I. Teshima and M. Sakamoto, 1985. Effects of dietary lipids, fatty acids and phospholipids on growth and survival of prawn (*Penaeus japonicas*) larvae. Aquaculture, 50: 39-49.

Knuckey, R.M., G.L. Semmens, R.J. Mayer and M.A. Rimmer, 2005. Development of an optimal microalgal diet for the culture of the calanoid copepod *Acartia sinjiensis*: Effect of algal species and feed concentration on copepod development. Aquaculture, 249: 339-351.

Koski, M., W.K. Breteler and N. Schogt, 1998. Effect of food quality on rate of growth and development of the pelagic copepod *Pseudocalanus elongates* (Copepoda, Calanoida). Mar. Ecol. Prog. Ser., 170: 169-187.

Koven, W.M., A. Tandler, G.W. Kissil and D. Sklan, 1992. The importance of n3 highly unsaturated fatty acids for growth in larval *Sparus aurata* and their effect on survival, lipid composition and size distribution. Aquaculture, 104: 91-104.

Kraul, S., A. Nelson, K. Brittain and D. Wenzel, 1988. Feeding and handling requirements for hatchery culture of the mahimahi *Coryphaena hippurus*. Quart. Univ. Hawaii Sea Grant Program, 10: 1-6.

Kraul, S., H. Ako, A. Nelson, K. Brittain and A. Ogasawara, 1992. Evaluation of live feeds for larval and postlarval mahimahi, *Coryphaena hippurus*. J. World Aquacult. Soc., 23: 299-306.

Leger, P., D.A. Bengtson, K.L. Simpson and P. Sorgeloos, 1986. The use nutritional value of Artemia as a food source. Oceanogr. Mar. Biol. Annu. Rev., 24: 521-623.

Li, P., K. Mai, J. Trushenski and G. Wu, 2009. New developments in fish amino acid nutrition: towards functional and environmentally oriented aquafeeds. Amino Acids, 37: 43-53.

Lowry, O.H., N.J. Rosebrough, A.L. Farr and R.J. Randall, 1951. Protein measurement with the Folin phenol reagent. J. Biol. Chem., 193: 265-275.

Lu, J. and T. Takeuchi, 2004. Spawning and egg quality of the tilapia *Oreochromis niloticus* fed solely on raw *Spirulina* throughout three generations. Aquaculture, 234: 625-640.

Mazorra, C., M. Bruce, J.G. Bell, A. Davie and E. Alorend *et al*., 2003. Dietary lipid enhancement of broodstock reproductive performance and egg and larval quality in Atlantic halibut (*Hippoglossus hippoglossus*). Aquaculture, 227: 21-33.

Mustafa, G.M. and H. Nakagawa, 1995. A review: Dietary benefits of algae as an addittive in fish feed. The Israeli J. Aquacult. Bamidgeh, 47: 155-162.

Nakagawa, H., S. Kasahara and T. Sugiyama, 1987. Effect of *Ulva* meal supplementation on lipid metabolism of black sea bream, *Acanthopagrus schlegeli* (Bleeker). Aquaculture, 62: 109-121.

Nandeesha, M.C., S.S. de Silva and D. Krishnamurty, 1993. Evaluation of Mixed Feeding Schedules in Two Indian Major Carps. *Catla* (*Catla calta*) and Rohu (*Labeo rohita*). In: Fish Nutrition in Practice, Kaushik, S.J.J. and P. Lequet (Eds.). Institute National De-la Recherche Agronomique, Paris, France, pp: 753-765.

Nanton, D.A. and J.D. Castell, 1998. Preliminary trials using a harpacticoid copepod, *Tisbe* sp., as a diet for larval haddock and American plaice. Bull. Aquacult. Assoc. Canada, 4: 38-40.

Nichols, P.D., J.B. Guckert and D.C. White, 1986. Determination of monosaturated fatty acid double-bond position and geometry for microbial monocultures andcomplex consortia by capillary GC-MS of their dimethyl disulphide adducts. J. Microbiol. Methods, 5: 49-55.

Phang, S.M., M.S. Miah, B.G. Yeoh and M.A. Hashim, 2000. *Spirulina* cultivation in digested sago starch factory wastewater. J. Applied Phycol., 12: 395-400.

Pillay, K.K. and N.B. Nair, 1973. Observations on the biochemical changes in gonads and other organs of *Uca annulipes, Portunus pelagicus* and *Metapenaeus affinis* (Decapoda, Crustacea) during reproductive cycle. Mar. Biol., 18: 167-196.

Rainuzzo, J.R., K.I. Reitan, L. Jurgensen, and Y. Olsen, 1994. Lipid composition in turbot larvae fed live feed cultured by emulsions of different lipid classes. Comparat. Biochem. Physiol. A Physiol., 107: 699-710.

Ravi, J. and K.V. Devaraj, 1991. Quantitative essential amino acid requirements for growth of catla, *Catla catla* (Hamilton). Aquaculture, 96: 281-291.

Reitan, K.I., J.R. Rainuzzo, G. Oie and Y. Olsen, 1997. A review of the nutritional effects of algae in marine fish larvae. Aquaculture, 155: 207-221.

Rey, C., R. Harris, X. Irigoien, R. Head and F. 2001. Influence of algal diet on growth and ingestion of *Calanus helgolandicus* nauplii. Mar. Ecol. Prog. Ser., 216: 151-165.

Roe, J.H., 1955. The determination of sugar in blood and spinal fluid with anthrone reagent. J. Biol. Chem., 212: 335-343.

Romdhane, M.S., B. Devresse, P. Leger and P. Sorgeloos, 1995. Effects of feeding (ù-3) HUFA-enriched *Artemia* during a progressively increasing period on the larviculture of freshwater prawns. Aquacult. Int., 3: 236-242.

Sargent, J.R., L.A. McEvoy and J.G. Bell, 1997. Requirements, presentation and sources of polyunsaturated fatty acids in marine fish larval feeds. Aquaculture, 155: 117-127.

Shelef, G. and C.J. Soeder, 1980. Algal Biomass Production and Use. 1st Edn., Biomedical Press, Elsevier, North Holland.

Shields, R.J., J.G. Bell, F.S. Luizi, B. Gara, N.R. Bromage and J.R. Sargent, 1999. Natural copepods are superior to enriched *Artemia* nauplii as feed for halibut larvae (*Hippoglossus hippoglossus*) in terms of survival, pigmentation and retinal morphology: relation to dietary essential fatty acids. J. Nutr., 129: 1186-1194.

Simon, C.J., 2009. Identi?cation of digestible carbohydrate sources for inclusion in formulated diets for juvenile spiny lobsters, *Jasus edwardsii*. Aquaculture, 290: 275-282.

Skalli, A. and J.H. Robin, 2004. Requirement of n-3 long chain polyunsaturated fatty acids for European sea bass (*Dicentrarchus labrax*) juveniles: Growth and fatty acid composition. Aquaculture, 240: 399-415.

Sorgeloos, P., 1980. The use of the brine shrimp *Artemia* in aquaculture. Proceedings of the International Symposium on the Brine Shrimp Artemia Salina. August 20-23, 1979, Christi, Texas, USA., pp: 25-46.

Sorgeloos, P., P. Lavens, P. Leger and W. Tackaert, 1991. State of the art in larviculture of fish and shellfish. Proceedings of the International Symposium on Fish and Crustacean Larviculture, August 27-30, 1991, Gent, Belgium, pp: 3-5.

Sorgeloos, P. and Ph. Leger, 1992. Improved larviculture outputs of marine fish, shrimp and prawn. J. World Aquacult. Soc., 23: 251-264.

Stottrup, J.G. and J. Jensen, 1990. Influence of algal diet on feeding and egg-production of the calanoid copepod *Acartia tonsa* Dana. J. Exp. Mar. Biol. Ecol., 141: 87-105.

Stottrup, J.G. and L.A. McEvoy, 2003. Live Feeds in Marine Aquaculture. Blackwell Science Ltd., Oxford, UK., Pages: 318.

Takeuchi, T., J. Lu, G. Yoshizaki and Y.S. Satoh, 2002. Effect on the growth and body composition of Juvenile tilapia *Oreochromis niloticus* fed raw *Spirulina*. Fish. Sci., 68: 34-40.

Tang, Y. and T.L. Hwang, 1966. Evaluation of the relative suitability of various groups of algae as food of milkfish in brackish-water ponds. FAO Fish. Rep., 3: 601-603.

Tocher, D.R., J.G. Bell, J.D. Castell and J.R. Sargent, 1994. Effects of purified diets containing different combinations of arachidonic and docosahexaenoic acid on survival, growth and fatty acid composition of juvenile turbot (*Scophthalmus maximus*). Aquaculture, 128: 315-333.

Venkat, H.K., N.P. Sahu and K.K. Jain, 2004. Effect of feeding Lactobacillus-based probiotics on the gut microflora, growth and survival of postlarvae of *Macrobrachium rosenbergii* (de Man). Aquacult. Res., 35: 501-507.

Venkataraman, S., 1972. Algal Biofertilizers for Rice Cultivation. Today and Tomorrow Publisher, New Delhi, pp: 75.

Watanabe, T., 1993. Importance of docosahexaenoic acid in marine larval fish. J. World Aquacult. Soc., 24: 152-161.

Witt, U., G. Quantz, D. Kuhlmann and G. Kattner, 1984. Survival and growth of turbot larvae *Scophthalmus maximus* L. Reared on different food organisms with special regard to long-chain polyunsaturated fatty acids. Aquacult. Eng., 3: 177-190.