UTILIZATION OF WATER SPINACH *IPOMOEA AQUATICA* LEAF MEAL AS PROTEIN SOURCE IN THE FEED OF *MACROBRACHIUM ROSENBERGII* POST LARVAE

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PANANGAD-KOCHI

DEDICATED TO

MY FAMILY

&

THRESIAMMA MAAM

DECLARATION

I hereby declare that this thesis entitled **"UTILIZATION OF WATER SPINACH *IPOMOEA AQUATICA* LEAF MEAL AS PROTEIN SOURCE IN THE FEED OF *MACROBRACHIUM ROSENBERGII* POST LARVAE."** is a bonafide record of research work done by me during the course of research and that the thesis has not previously formed the basis for the award to me of any degree, diploma, associateship, fellowship or other similar title of any other University or Society.

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CERTIFICATE

Certified that this thesis entitled "**UTILIZATION OF WATER SPINACH *IPOMOEA AQUATICA* LEAF MEAL AS PROTEIN SOURCE IN THE FEED OF *MACROBRACHIUM ROSENBERGII* POST LARVAE**" is a record of research work done independently by **Miss.Sijimol.A.P** under my guidance and supervision and that it has not previously formed the basis for the award of any degree, fellowship or associateship to her.

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INTRODUCTION

INTRODUCTION

Aquaculture, probably the fastest growing food-producing sector, now accounts for almost 50% of the world's food fish and is perceived as having the greatest potential to meet the growing demand for aquatic food. Capture fishery production in India both in the marine as well as freshwater sector is being increasingly plateaued. While the annual growth rate of capture fishery has been somewhat stagnant, aquaculture production has been recording a phenomenal, annual growth of 9% (New, 1996).

Freshwater prawns of the Genus *Macrobrachium* are distributed throughout the tropical and subtropical regions and more than 100 species are known to exist today (New, 2000). However the most important species among them is the giant fresh water prawn, *Macrobrachium rosenbergii*. This is the best species for aquaculture since it grows well in both freshwater and brackish waters. The favorable attributes for the farming of giant freshwater prawn are its successful reproduction in captivity, established technologies for larval rearing, faster growth rate, high tolerance to wide ranges of salinity and temperature, omnivorous feeding habit, compatibility in polyculture, absence of major disease problems, wider consumer acceptability, high market value, tolerance to water quality changes, ability to cope with handling stress and ability to feed on unconventional feeds (El-Sayed,1997;Ayyappan and Pillai,2005).

The farming of the giant freshwater prawn *Macrobrachium rosenbergii*, popularly known as 'Scampi' has been expanding in India for the past 10 years. In its natural environment the freshwater prawn is able to satisfy its particular nutrient requirements from a variety of sources, which includes aquatic worms, insects, small molluscs, crustaceans, plankton and organic detritus (Mitra *et al.*, 2005). However, under controlled conditions of culture, it will become necessary to provide the prawn with appropriate levels of nutritionally balanced feeds. Effective formulation of a balanced diet for a particular species requires a detailed understanding of the nutritional requirements of the species. In intensive culture systems, feed represents the largest input and feed cost often accounts for 40-60% of the

operational cost (DeSilva and Davy, 1990; Piedad-pascual *et al.*, 1990; Akiyama *et al.*, 1992; Sarac *et al.*, 1993), and may rise to as high as 75% (Shang, 1981).

Profitable fish culture requires a steady supply of formulated feed in which proteins serve as both growth nutrient and energy currency. Thus, formulation of low cost feeds using the cheapest sources of protein is essential to hasten fish production and to reduce feed cost (Krishnankutty, 2005).

The cost of the feed mainly depends upon the cost of the protein source. The most commonly used protein source in fish/prawn feeds is fish meal. Owing to the high cost and fluctuating quality as well as uncertain availability there is shortage of the above protein sources, which has led to their price escalation in the market (Alceste, 2000). Hence, many feed ingredients alternative to fish meal at varying levels are being sought to attain sustainable aquaculture in the current millennium. Considerable emphasis has been focused on the use of conventional plant protein sources, such as soyabean, ground nut, cotton seed and rape seed meal (Jackson *et al.*, 1982; El-Sayed, 1990). However their scarcity and competition from other sectors for such conventional crops for livestock and human consumption as well as industrial use, make their cost too high and put them far beyond the reach of fish farmers or producers of aquafeeds (Fasakin *et al.*, 1999). Therefore, in order to attain a more economically sustainable, environment friendly and viable production, research interest has been directed towards the evaluation and use of unconventional protein sources particularly from plant products such as seeds, leaves, agricultural byproducts and aquatic weeds (Olvera-Novoa *et al.*, 1988; El-Sayed, 1999; Siddhuraju and Becker, 2001). Associated with the development of intensive farming of *Macrobrachium rosenbergii*, the development of production of diets using local and cheap ingredient is felt inevitable.

The aquatic weed menace is reaching alarming proportions in many parts of the country. Their physical removal will become economical if monetary return from harvested plants can be found through their commercial use (Patnaik *et al.*, 1991; Mohanty and Das, 1995; Ray and Das, 1995; Agbede and Falaye, 1997; Guru and Patra, 2007; Mishra, 2007). In

this context, attention has to be paid to utilize protein rich aquatic plant leaves as feed ingredients.

Although attempts have been made to use the leaves of aquatic weeds in fish feeds, there is no information regarding the utilization of water spinach leaves in fish/prawn feeds. Therefore the present study is aimed at utilizing the leaves of *Ipomoea aquatica* (water spinach), the ubiquitous aquatic weed, as a protein source in the feed of *Macrobrachium rosenbergii* post larvae.

REVIEW OF LITERATURE

2. REVIEW OF LITERATURE

2.1. NUTRITIONAL REQUIREMENTS OF FRESHWATER PRAWNS

Successful and sustainable farming of the giant freshwater prawn depends up on the application of nutritionally adequate, eco-friendly and economically viable supplementary feeds. Feed is a major operational input and costs normally range from 30-50% of total operational expenditure (Sultan, 2005). In view of this, supplementary feeds should be scientifically formulated, optimally processed and judiciously applied, depending upon specific nutritional needs and intensity of farming operations. Ingredients for prawn feeds are of animal as well as plant origin. Several locally available feed ingredients have been evaluated as the cost effective feed ingredients for prawn. Mixed feeding schedules including alternate high and low protein diets have resulted in saving of feed cost with out affecting the growth and survival (Naik and Murthy, 2001; Sultan, 2005). Feeding studies carried out on *M. rosenbergii* are mostly concentrated on larval and juvenile stages (Ling 1969; James, 1990; Sherief, 1992; Daniels *et al.*, 2000).

2.1.1. Protein

Protein, being a dietary constituent of major importance, influences the growth and production of prawns (Antiporda, 1986). The source, dietary level and amino acid composition of protein in relation to shrimp and prawn nutrition has received much attention, because it is the largest and most expensive component in the feed (New,1976). As for the nursery phase of *Macrobrachium rosenbergii* is concerned, the quantitative dietary protein requirement is still in considerable uncertainty. The estimated dietary protein requirement for *Macrobrachium rosenbergii* post larvae and juvenile range between 30 to 35 % of crude protein (D' Abramo and New, 2000). Starter diets usually contain 32% while grower and finisher diets have a lesser level of 30% protein. Several studies showed that better growth performance of *Macrobrachium rosenbergii* between 30 and 40% protein levels (Balazs and Ross, 1976; Millikin *et al.*, 1980; D' Abramo and Reed, 1988; Gomez *et al.*, 1988;

AQUACOP, 1990; Law *et al.*, 1992; Koshio *et al.* , 1992). Fruechtenicht (1988) found better performance of *Macrobrachium rosenbergii* in 51.25% protein diet. But Gomez *et al.* (1988) reported that dietary protein level as low as 13 % was sufficient for *Macrobrachium rosenbergii* and the ratio of protein: starch of 1:1 leads to good growth and high survival rate, although feed efficiency was the highest at the protein to starch ratio of 1:3. However, protein efficiency ratio was the highest for the diet of 384 Kcal /100 g diet. Recent studies pointed out that higher dietary protein levels neither result in higher freshwater prawn yields nor significantly influence their survival rate, harvest size, yield or gross FCR (Posadas *et al.*, 2001).

If protein in the diet is insufficient, the already stored protein is withdrawn from the tissues to carry on the vital life functions, thereby resulting in rapid reduction in growth (Paulraj, 1993). Millikin *et al.* (1980) reported better growth, FCR and PER in *Macrobrachium rosenbergii* fed 40% protein diet and observed decreased growth rates in 49% protein diet. The elevated rates of protein catabolism were found to be the reason for decreased growth in higher protein level. Frechienicht (1988) reported better growth of prawns at 30.3 and 51.2% protein levels in the diet. Law *et al.* (1992) recorded maximum growth in post larvae of *Macrobrachium rosenbergii* when fed with 40% protein diet as against 25, 30 and 50% protein levels. Utilization of dietary proteins for energy production and metabolism is both nutritionally and economically wasteful. It is necessary to spare protein for growth by optimizing the level of non-protein energy sources (Alava and Pascual, 1987). Gomez *et al.* (1988) reported that high starch content promote PER suggesting that dietary protein was spared by supplementation of starch in *Macrobrachium rosenbergii*. In case the energy derived from carbohydrate and lipid is insufficient, prawn metabolizes protein for energy requirement which is uneconomical and ammonia produced in deamination process can be toxic to prawn. Hence, the optimum levels of protein in the feed is essential and 30-35% protein is suitable for *Macrobrachium rosenbergii* feed with total energy of 350Kcal GE /100g feed (El-Sayed ,1997). Felix and Jayaseelan (2006) revealed the need for 40% protein diet to achieve better growth performance in *Macrobrachium rosenbergii* in nursery phase.

They found that protein sparing could be maximized at L: CHO of 1:3.7 to achieve better growth of *Macrobrachium rosenbergii* and efficient utilization of feed.

Varying levels of proteins ranging from 35-45% (Ang *et al.*, 1992; Das *et al.*, 1993; 1996) have been considered as ideal for brood stock of fresh water prawn under experimental conditions. Similarly D' Abramo *et al.* (1995) had reported a successful use of a high energy, high protein (40%) salmonid diet for brood stock of *Macrobrachium rosenbergii.* Das *et al.* (1996) evaluated the efficiency of 12 formulated diets on the reproductive performance of *Macrobrachium rosenbergii* and recommended that a brood stock diet with 40% crude protein and a gross energy level of 400Kcal/100g diet was ideal. Based on biochemical characterization of the tissues of *Macrobrachium rosenbergii,* a lower level of 30% CP has been considered to be sufficient to meet the requirements of pond reared brood stock (Bindu *et al.*, 2001). Mitra *et al.* (2005) reviewed that a protein level of 38-40% and energy level of 3.7-4 Kcal/g feed are optimal for *Macrobrachium rosenbergii* brood stock. Additional information provided by the same authors suggests that brood stock reared in ponds having natural food requires only about 30% protein in their diet.

The protein content of commercial prawn feeds have been reported as 23.8 to 38.5 % in Hawaii (Corbin *et al.*, 1983), 28.36 % in Taiwan (Hsieh *et al.*, 1989) and 25-30% in French Guiana (IFREMER.1989). Clifford and Brick (1979) reported that optimum growth of prawns was achieved with diet having 25% protein and 1:4 lipid: CHO ratio. According to New (1976) the optimal level of dietary protein for different species of prawns is between 27% and 35%. In case of juvenile *Macrobrachium rosenbergii* the requirement may be somewhat higher. According to Behanan and Mathew (1995) a diet containing 30-40 % protein produces better growth in *Macrobrachium rosenbergii.* Boonyaratpalin and New (1980) suggested that desirable protein level of feed from economical stand point was 15%, at least for the first four months of rearing. Ravishankar and Keshavanath (1988) found that *Macrobrachium rosenbergii* utilized feed pellets containing silkworm pupae and shrimp waste more efficiently and gave higher specific growth rate than diet having fish meal, silkworm pupae alone or silk worm pupae plus clam meat. Unnikrishnan *et al.*(1991) were able to substitute extracted silk

worm pupae for extracted clam meat as semi purified diet for *Macrobrachium rosenbergii* post larvae without any detrimental effect on the survival ,growth rate and protein efficiency ratio.

Behanan *et al.*(1992) conducted another experiment on the post larvae of *Macrobrachium rosenbergii* by using pelleted feeds with 33-44% protein and found that the diet containing 33% protein was ideal. James *et al.* (1990) found that *Spirulina fusiformis* can be used as a supplementary protein but cannot be secured as a sole protein source in *Macrobrachium rosenbergii* post larvae. Good growth and survival have been obtained by feeding with proteins having amino acid profile similar to the tissues of shrimp itself (Kanazawa, 1992). Pezzato *et al.* (1995) evaluated the influence of three protein levels (30, 40 and 50%) in the diet of post larvae of *Macrobrachium rosenbergii*. Higher protein levels in the diet were needed by in the first 10 weeks and after 20 weeks higher weight gain were obtained with a diet containing 41.38 % crude protein.

A point worth mentioning is that no study has hitherto evaluated the protein requirement of freshwater prawn in well fertilized ponds having adequate natural food in the form of plankton or benthic micro and macro fauna. In fact, the quality of protein, which may be actually utilized by the animals in well fertilized and productive ponds environments, is not known. In the absence of such published information on the nutrient requirements of fresh water prawn in pond based farming systems, it is to be believed that almost all the commercial aquafeeds may be over formulated so far as protein is concerned. This justifies the concern for a revalidation of the protein requirement of post larvae and juvenile freshwater prawn to be carried out under practical farming conditions. Knowing that the species shows good survival and growth on low protein (15-20%) diets in well fertilized ponds decreasing the current protein levels by 10-15% at the initial stages of farming could certainly help farmers reduce the operating costs and achieve better margins of production (Raghavan and Prasad,2006).

2.1.2. Amino acids

Using radio isotopic analysis, Cowey and Forster (1971) and Shewhart *et al.* (1972) showed that arginine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tryptophan, tyrosine and valine are essential for *Palaeomon serratus and Penaeus aztecus* respectively. The prawn requires the same ten essential amino acids as other crustaceans and fish species, but quantitative requirements have not been determined. Watanabe (1975) and Watanabe *et al.* (1976) have shown that tyrosine is essential for *Macrobrachium rosenbergii* in addition to histidine, methionine, leucine, isoleucine, valine, tryptophan and phenylalanine. A conspicuous variation in *Macrobrachium rosenbergii* is that in contrast to other crustaceans studied so far, lysine seems to be non-essential for *Macrobrachium rosenbergii* (Watanabe, 1975). Similar results were obtained for *Macrobrachium ohione* by Miyajima *et al.* (1976) except for tryptophan. Diets based on essential amino acid profile of *Macrobrachium* egg that would yield high fecundity and high hatching contained 40 % protein and 400 Kcal GE/100 g diet. The essential amino acids for shrimps and prawns are qualitatively similar to those of other animals (New, 1976). Similar observations by Stahl and Ahearn (1978) had shown that lysine is not essential for growth and survival of *Macrobrachium rosenbergii.* These authors also investigated the essentiality of arginine,histidine and tryptophan, and reported that these three amino acids are found to be non-essential for *Macrobrachium rosenbergii* ,but cautioned that gut and /or tank bacteria may have supported the prawn with the above amino acids in sufficient amounts for growth.

Smith *et al.* (1987) reported the incapability of *Macrobrachium rosenbergii* in synthesizing asparagines, which is regarded as another essential amino acid. This requires further investigation regarding its categorization. The whole body amino acid profile of prawn can be indicative of its quantitative as well as qualitative amino acid requirement and the ratio of total amino acid to essential amino acid is similar for *Macrobrachium rosenbergii.* Dall and Smith (1987) reported that dietary sources of amino acids regarded as non-essential may be required if they have high rate of metabolic turnover. Alanine is thought to have an

osmoregulatory function, while taurine is involved in cardiac and neural processes in *Macrobrachium rosenbergii* (Smith *et al.*, 1987). Tidwell *et al.* (1993) had shown that replacement of fish meal with soyabean and distillers by-products in diets for *Macrobrachium rosenbergii* caused significant increase in concentration of glutamine, proline, alanine, leucine and phenylalanine.

Kanazawa (1992) found that methionine enriched soyabean protein was preferable to crystalline methionine when used to supplement a methionine deficient diet. Attempts to support amino acid deficient diets with crystalline amino acid or basal diet consisting of crystalline amino acid have not been successful with shrimp (Lovell, 1998).

The amino acid composition of the prawn muscle is used to provide guidance values in feed formulation (Mitra *et al.*, 2005). The effects of utilization of feed with balanced amino acid or poor quality protein are manifested in slow growth, low protein efficiency and higher dietary protein requirement.

2.1.3. Lipids and fatty acids

Lipids are important in the diet of prawns, next to proteins. Besides acting as an energy source, it has been found that dietary lipids have the highest protein sparing action. Unlike fishes, crustaceans cannot tolerate high levels of dietary lipid (Sheen and D'Abramo, 1991). In comparison to penaeid shrimps, *Macrobrachium rosenbergii* requires comparatively low lipid levels in diets. Joseph and Williams (1975) reported best growth in post larvae of *M. rosenbergii* fed on diets with 35 shrimp head oil. Hilton *et al.* (1984) indicated that the total lipid content in the diet should not exceed 10% for *M.rosenbergii*. Sheen and D'Abramo (1991) found that 6% inclusion rate was optimal, while 0, 10 and 12% levels depressed growth in *M.rosenbergii* juveniles. They also observed significant reduction in weight of *M.rosenbergii* juveniles when 10-12 % of the same lipid sources were used and when a 2:1 mixture of cod liver oil and corn oil was fed at dietary levels ranging from 2-10%, weight gain of juvenile *Macrobrachium rosenbergii* was not significantly different and dietary lipid levels

which exceed 10% are generally associated with a reduction in growth rate possibly due to the inability to process such high levels. Fox *et al.* (1994) reported that inclusion of more than 10% lipid in prawn diet frequently led to suboptimal growth.

Crustaceans are known to synthesize certain groups of fatty acids *de novo* in their body, viz., palmitic (16:1), stearic (18:0) and oleic (18:1) acids (Kanazawa *et al.*, 1979). Sandifer and Joseph (1976) demonstrated better growth of juvenile *Macrobrachium rosenbergii* ,fed with diets supplemented with 3% shrimp head oil rich in 20:5n-3 and 22:6n-3. Castell (1983) noted that whole body tissue of *Macrobrachium rosenbergii* contained higher level of n-6 Poly unsaturated fatty acids (PUFA) and that maximum dietary 20:4n6 requirement was 0.075%. Reigh and Stickney (1989) reported that the growth was negatively affected in *Macrobrachium rosenbergii* feeding on the diet with 18:3n3 as the only source of dietary lipid, as *Macrobrachium rosenbergii* are incapable of synthesizing neither 18:2n6 nor 18:3n3. Teshima *et al.* (1992) found that *Macrobrachium* species are incapable of elongating and desaturating 20:5n3 to 22:6n3 but are able to convert 22:6n3 to 20:5n3. However the ability for synthesis of long chain fatty acids with double bonds is absent or very limited in crustaceans and fresh water prawn requires dietary HUFAs for better growth (D'Abramo and Sheen1993). Teshima *et al.* (1994) found that optimal n-6 to n-3 fatty acid ratio was 20:1, but optimal ratio of 18:2n6 and 18:3n3 has not been established. Nutrition studies indicated that 5-9 % of a mixture of fish oil and soyabean at 1:1 ratio give best growth, FCR and PER.

Roustainan *et al.* (2001) reported that *Macrobrachium rosenbergii* are capable of elongating and desaturating C16:0 to C18:0, C18:2n6 to C20:4n6, C18:3n3 to C20:5n3. Both n-3 and n-6 HUFAs at dietary levels of 0.075% are known to increase weight gain and feed efficiency remarkabably, in addition 18:2n-6 and 18:3n-3 is also required (Mitra *et al.*, 2005).

2.1.4. Sterols

Many species of fresh water and marine crustaceans have been shown to require an exogenous source of sterols for normal growth and development (Kanazawa, 1982; 1985). *Macrobrachium rosenbergii*, like other crustaceans, is unable to synthesize cholesterol due to

the absence of the enzyme 3 hydroxy 3methylglutaryl CoA redutase, but can convert betastiosterol to cholesterol. Therefore, there is need to incorporate cholesterol in diet. The dietary cholesterol requirement varies from 0.12 % (Briggs *et al.*, 1988) to 0.5-0.6 % (Sherief *et al.*, 1992). The dietary requirement of cholesterol in *Macrobrachium rosenbergii* is approximately 0.3-0.6 % in diet. Substitution with 0.6 % ergosterol or stigmesterol is generally not so effective compared to 0.6% cholesterol. However, a mixture of phytosterols has been found to be as effective as cholesterol. So like in penaeid shrimp feed, there is no need to add high levels of purified cholesterol in fresh water feeds provided the ingredients contain sufficient levels of phytosterols (Mitra,*et al.*,2005).

The freshwater prawn has no fixed dietary lipid requirement (D'Abramo and New, 2000). Bindu *et al.* (2001) reported that the total lipid content in hepatopancreas and ovary and the total cholesterol content in hepatopancreas and eggs were comparatively lower in pond reared *Macrobrachium rosenbergii* than the wild caught ones. This study further suggests that dietary components like fatty acids and cholesterols are needed by the brood stock at higher levels than that are normally incorporated in grow out feeds. Saturated and monounsaturated fatty acids were greater than poly unsaturated fatty acids in *Macrobrachium rosenbergii* eggs (Cavalli *et al.*,2000), newly hatched larvae (Roustaian *et al.*,2001) and the midgut and ovary of matured females (Cavalli *et al.*,2000). Hien *et al.* (2001) suggested that the quality of the lipid source used in brood stock feeds clearly affected the fecundity, egg fatty acid composition and offspring quality in *Macrobrachium rosenbergii.* The decosahexanoic acid (DHA) is the most utilized individual fatty acid during embryonic development of *Macrobrachium rosenbergii* (Clarke *et al.*, 1990). The significance of n-3 HUFA in brood stock diets and their relation to egg hatching and significance of 18:2n-6 in relation to fecundity is well documented in *Macrobrachium rosenbergii* (Cavalli *et al.*, 1999; D'Abramo and New, 2000).

The literature on post-larval lipid nutrition suggests that *Macrobrachium rosenbergii* is benefited from an n-3 HUFA enriched diet. The vitellogenin titre of fresh water prawn given diet containing high HUFA was higher than those of prawn fed on diets containing low HUFA

and trash fish (Hien and Phu, 2000). Cholesterol is critically involved in the reproductive process of *Macrobrachium rosenbergii* (Cavalli *et al.*, 2001).

2.1.5. Phospholipids

Hilton *et al.* (1984) reported that supplemental lecithin was required by *Macrobrachium rosenbergii* fed a semi- purified diet containing casein- gelatin- wheat gluten (18:1:1) as protein source. No significant difference was detected in either weight gain or mortality of the prawn fed diets containing levels of refined soyabean lecithin ranging from 0 % to 10 % for 12 weeks. No sign of deficiency or moult death syndrome was observed in any of the specimens. It is established that the supplementation of 1 and 2% soyabean lecithin to either casein or crab protein based diets does not improve the percentage weight gain and survival of *Macrobrachium rosenbergii* (Kanazawa, 1993).

Contradictory results were reported by Koshio *et al.* (1994) who noted that phospholipids had no positive effect on growth and feed conversion ratio when prawns were reared in groups; but the effect was observed when reared individually. Tiwari and Sahu (1999) reported 2% soya lecithin gave growth and survival than control and 5% soya lecithin inclusion along with 0.5% ground nut oil and 0.5% cod liver oil provided maximum growth for juveniles.

The freshwater prawn also has limited ability to biosynthesize phospholipids *de novo*. Manipulation of the level of phospholipids in brood stock had no profound effect on the fecundity, egg size,hatchability,starved larval size or larval quality (Cavalli *et al.*,2000) indicating that fresh water prawn do not need supplementary phospholipids and are satisfied with the phospholipids containing ingredients present in commercial diets. A basal level of 0.8% dietary phospholipids is required to meet the demand of the scampi brood stock (Mitra *et al.*, 2005).

2.1.6. Carbohydrates

Prawn can efficiently utilize carbohydrates, as it possesses enzymes that digest starch, cellulose and chitin. Its apparent digestibility of cellulose is about 80%. As the dietary cellulose increases, the amount of cellulase also increases while amylase reduces. Hence, cellulose serves as source of energy in *Macrobrachium rosenbergii* feed (Briggs, 1991; Del-Carmen-Gonzalez-penna *et al.*2001). The optimum carbohydrate to lipid ratio is 1:4. Complex polysaccharides including starch and dextrin are more effectively utilized than simple sugars.

Studies have also proved that complex dietary polysaccharides are utilized better by freshwater prawn and yield higher growth rates when compared to disaccharide and monosaccharide (Briggs, 1991). Carbohydrate activity for freshwater prawn is presumed to be comparatively higher than that of carnivorous species of Penaeid shrimps (D'Abramo and New, 2000). Anilkumar (1994) found that the protein level for *Macrobrachium rosenbergii* juveniles could be lowered from 35 to30% by increasing the carbohydrate level from 20 to 30% without affecting the growth, survival and feed efficiency. This suggests that a 5% reduction of protein can be achieved by enhancement of 10% carbohydrate, thus reducing the cost of the feeds with out sacrificing the nutritional quality of the formulated feed. Carbohydrates do not appear to be essential for crustacean brood stock diets.

The comparatively high specific activity of amylase found for *Macrobrachium rosenbergii* supporting the fact that the species efficiently utilizes carbohydrate as a source of energy (Mitra *et al.*, 2005). As of now, little attention has been paid to the incorporation of high levels of carbohydrate source into freshwater prawn feeds. Thus there is plenty of scope for reducing both feed costs and pollution by incorporating cheap, locally available and digestible carbohydrate sources in fresh water prawn diets (Raghavan and Prasad, 2006). A perusal of the literature reveals the absence of any published information on the carbohydrate nutrition of freshwater prawn brood stock.

2.1.7. Fibre

Gomez *et al.* (1988) reported poor growth and depressed locomotive activity in *Macrobrachium* fed on diet containing 20% cellulose. Little is known about the nutritional significance of fibre in the diet of crustaceans. It has been a common practice among crustacean nutritionist to include cellulose (primarily as a dietary filler) in experimental rations at levels ranging from 0 to as high as 75% of the total dry matter (New,1990). Zimmerman *et al.*(1992) incorporated four inexpensive forages (the legume *Clitoria ternalea* and grasses *Bacharia purpurescens, B. arrecta and B.humidicola*) as fibre sources, which constituted 75% of the fibre content of the diets and found that the diet containing *B. purpurescens* gave highest average individual weight and biomass in *Macrobrachium rosenbergii.* Growth, feed efficiency and protein efficiency ratio improve when dietary fibre increases from 0.4 to 0.8 %. The prawns are known to utilize as high as 30% dietary fibre (Mitra *et al.*, 2005).

2.1.8. Vitamins

Harrison (1997) indicated that vitamin D is important in crustacean brood stock diets due to its probable role in calcium and phosphorous metabolism. A study by Cavalli *et al.* (2001) described the essentiality of vitamins C and E during maturation and reproduction in *Macrobrachium rosenbergii.* Levels of 60 mg ascorbic acid and 300mg of tocopherol/kg of feed are considered sufficient for proper reproduction and offspring viability in prawn brood stock (Mirta *et al.*, 2005). The same authors also suggested that, feeding female prawn with higher levels of both these vitamins (each around 900 mg/kg) might improve larval quality including higher tolerance to ammonia stress. It has also been reported that vitamin E at 200mg/kg diet modulated the antioxidant defense system by decreasing lipid peroxidation in the hepatopancreas. Recent evidence suggests that vitamin C with a dosage 0.15 % of body weight is optimum to accelerate gonadal maturation in fresh water prawn (Pamungkas *et al.*, 2005).

Macrobrachium species feeds on almost every thing on the pond bottom and all the time. Therefore, vitamin deficiency symptoms rarely occur. For *Macrobrachium rosenbergii*, high mortality is the only deficiency sign reported for vitamins. D' Abramo *et al.*(1994) who estimated the dietary vitamin C requirement to be approximately 100 mg/kg, a level that is consistent with reported vitamin C requirement of species of marine shrimp. Some crustacean have a limited ability to synthesize ascorbic acid (Lighter *et al.*, 1979), but this is considered insufficient to meet metabolic requirements, as most species tested to date require a dietary source of ascorbic acid.

Cavalli *et al.* (2001) found that a high correlation between the deposition of lipid and α-tocopherol in the ovary of *Macrobrachium rosenbergii* and concluded that this was in line with the antioxidative function of this vitamin. In comparison to other tissues, the muscles had relatively lower concentrations of ascorbic acid and α-tocopherol, but as the muscle mass comprised around 40% of the live weight of female prawns, it represented the most important storage site of these vitamins in *Macrobrachium rosenbergii*. Therefore, from a metabolic viewpoint, it seems more reasonable that ascorbic acid could be consumed during embryonic development, as previously suggested for *Macrobrachium rosenbergii.*

Akiyama *et al.* (1992) recommended 50mg /kg for practical commercial shrimp feeds. Ittoop (1996) reported a maximum weight gain in *Macrobrachium rosenbergii* juveniles when fed on diet containing 150 mg of vitamin c in the form of CVC-F90 (Hydrogenated vegetable oil form of vitamin C) per kg dry weight. When alternate plant protein sources were used in diets containing the same concentration of digestible energy and protein and when the diets formulated meet the nutritional requirements of the animal being fed, similar performance may be expected (Cruz-Suarez *et al.*, 2001).

2.1.9. Minerals

Information on quantitative mineral requirement of fresh water prawn is limited. Harrison (1990) had suggested that mineral deficiencies or imbalances could have negative

impact on crustacean reproduction with regard to oocyte resorption, reduction in reproductive performance and egg quality. Few studies have been conducted with the help of diets that were formulated with mineral mixes including calcium, phosphorous, magnesium, sodium, iron, manganese and selenium (Chamberlain 1988; Xu *et al.*, 1994; Marsden *et al.*, 1997; Mendoza *et al.*, 1997). There are no studies that have dealt with mineral nutrition associated with brood stock of *Macrobrachium rosenbergii.* Dietary supply of calcium seems to improve growth of freshwater prawn. Performances of prawn were better when calcium was provided at 3% level in soft water (calcium concentration at 5 ppm). Even when the calcium concentration was higher at 74 ppm, performance improved when calcium was provided at1.8%. The optimum level of zinc was at 50-90 mg/kg diet. Growth and feed conversion efficiency declined at higher dietary doses (>90mg/kg) of zinc (Mitra *et al.*, 2005).

Quantitative requirements of crustaceans for potassium have yet to be studied. With the exception of osmoregulation, physical functions of minerals in aquatic species appear to be similar to those in terrestrial animals (Lovel, 1989). Potassium (k) is the principal intracellular cation. For aquatic animals, the quantitative requirement for growth has been studied in only a few species of fish. For example 0.8/100g and 0.26g/100g diet have been reported as the requirements for chinook salmon and cat fish respectively (Shiav and Hsieh, 2001).

Prawns have neutral or weak acidic digestive tract; therefore, water soluble minerals are more utilizable. Researches in quantitative mineral requirements for *Macrobrachium rosenbergii* are limited. Dietary Zinc required for best growth and feed conversion is 90 mg/kg diet (Rath and Dube, 1994). Supplementation of calcium in diet was important when minerals in water are low. Dietary calcium required for best growth and survival is 0.3 to 1.8 % when calcium carbonate level in culture water was about 51-74 mg/l (Zimmermann *et al.*, 1994). The optimum Ca: P ratio was 1.5:1 in prawn feed. Further research should be carried out on mineral requirements in *Macrobrachium rosenbergii.*

2.2. PROTEIN SOURCES

A variety of proteins from plant and animal sources and the substitution effect of one source with another in different species of shrimp and prawns have been evaluated by several investigators with varying degree of success. Profitable fish culture requires unfailing supply of formulated fish feed in which proteins serve as both growth nutrient and energy currency. Thus, formulation of low-cost feeds using the cheapest sources of proteins is essential to hasten fish production and to reduce feed cost (Krishnankutty, 2005).

2.2.1. Animal protein sources

Various animal protein sources have been evaluated in the formulation of prawn feeds by several investigators, with varying degrees of success. Among the various animal protein sources of fresh water, marine and terrestrial origin crustacean and molluscan meals of marine origin were found to be better utilized by shrimps and prawns (Forster and Beard,1973; Deshimaru,1982; Gopal,1986; Sherief,1987; Kompaing,1990; Villarrel *et al.*,1991; Josekutty and Jose,1992; Cruz-Suarez *et al.*,1993; Anilkumar,1994; Dalfiah *et al.*,1998 ; Kabangnga *et al.*,1999).

Among the various crustacean meals, shrimp meal is one of the most commonly used protein source in the formulation of prawn feeds. Shrimp meal has high crude protein content and several essential amino acids (Forster, 1976). Besides it is a good source of fatty acids (Sandifer and Joseph, 1976) and appears promising for compounding shrimp and prawn feed (Venkataramiah *et al.*, 1978; Ali, 1998; Zimmermann *et al.*, 1991; Das *et al.*, 1995; Dalfiah *et al.*, 1998; Kabangnga *et al.*, 1999).

Zimmermann *et al.* (1991) compared the use of marine shrimp meal, fish meal and meat meal in 30% protein diets for post larvae of *Macrobrachium rosenbergii* and obtained best biomass production with the shrimp meal based diet compared to other two diets. Jayalakshmy and Natarajan (1994) reported highest production ,conversion efficiency and

lowest food conversion ratio for *Macrobrachium idella* fed on diet containing prawn waste than diets based on fish meal and clam meat. Das *et al.* (1995) observed the highest performance of prawn meal based diet for *macrobrachium malcomsonii* compared to mussel meat, fish meal and silk worm pupae based diets. Reena and Qureshi (1996) also reported superior performance of prawn meal based diet in terms of growth and food conversion in *Macrobrachium dayanum.*

In addition, the use of shrimp head waste in compounded ration also appears promising since it contains several essential amino acids which include high growth rate in prawns (Forster, 1976). It serves as a good source of fatty acids and pigments for use in prepared feeds for *Macrobrachium rosenbergii* (Sandifer and Joseph, 1976) and many marine animals (Joseph and Williams, 1975). However Law *et al.* (1990) reported lower digestibility of shrimp meal based diet than copra cake, soyabean meal and wheat flour based diets for *Macrobrachium rosenbergii.*

Crab protein has been found to be a good protein source for the growth and survival of crustaceans (Boghen *et al.*, 1982; D'Abramo and Reed, 1988; Freuchtenich *et al.*, 1988; Bordner, 1989; Reed and D'Abramo, 1989; Villarreal *et al.*, 1991; Koshio *et al.*, 1992). Feed formulation using protein concentrate from rock crab (*Cancer irroratus*) resulted in superior growth and survival of lobster *Homarus americanus*, when compared to several other proteins extracted from locally available marine organisms (Boghen *et al.*, 1982). However, Koshio *et al.* (1992) observed the superiority of dietary soyabean protein (SBP) over the crab protein for *Macrobrachium rosenbvergii* and reported that the better utilization of SBP may be done for removal of antinutritional factors during purification process. Raje and Joshi (1992) have also obtained higher efficiency of crab meal in combination with other protein sources in the diet.

Another potential crustacean protein source which is inexpensive and locally available is the stomatopod- *Oratosquilla neap*, the mantis shrimp and the use of squilla meal in the formulated feeds for *Macrobrachium rosenbergii* has been reported by Anilkumar (1994). Molluscs have been reported to be excellent feed for prawns (Sherief, 1989; Anilkumar,

1994). Minamizawa and Murizane (1970) obtained better results when *Artemia* nauplii were supplemented with chopped short necked clam for the larvae of *Macrobrachium rosenbergii*. Anilkumar (1994) found highest growth and food conversion efficiency in *Macrobrachium* juveniles fed on clam meat based diet in comparison to diets based on squilla, shrimp head waste and silk worm pupae as single protein sources. Contrary to this Jayalakshmy and Natarajan (1994) obtained poor food conversion ratio, production and food conversion efficiency of clam meat based diet compared to shrimp waste and fish meal as sole protein sources for *Macrobrachium idella*. Das *et al.* (1995) reported that for *Macrobrachium malcomsonii*, mussel meat based diet was found to be good but only second to the diet based on prawn meal.

Squid meal is another molluscan source of protein with high nutritive value. Although, it has been tried as a protein source in many penaeid shrimps (Deshimaru and Shigueno, 1972; Dokken and Lawrence, 1985; Cruz-Ricque et al., 1987), its use as a protein source in *Macrobrachium rosenbergii* was evaluated on by Anilkumar (1994).

Although, fish meal is one of the most common ingredients used in the commercial shrimp feeds and is a high quality source for fin fishes, it seems to have lower nutritive value for shrimps and prawns, especially when used as the sole protein source. The poor performance of fish meal in the latter case may be due to its inability to provide all essential amino acids, which are apparently essential for shrimps and prawns. Fish meal, in general is found to be poor source in threonine, alanine, phenylalanine, arginine and histidine (Lovel, 1989). While comparing different animal and plant protein sources, Sick and Beaty (1975) obtained poor growth rate in *Macrobrachium rosenbergii* fed on fish meal based diet which was lower than diet based on soyabean meal. Sherief (1989) also observed poor performance of *Macrobrachium rosenbergii* to fish meal based diet compared to clam meat. Das *et al.* (1995) reported that for *Macrobrachium rosenbergii* fish meal based diet produced lower growth rate, protein efficiency ratio and higher food conversion ratio than prawn meal and mussel meat based diets.

2.2.2. Plant protein sources

Prawn feeds based exclusively on plant protein sources have often produced poor results, since many of the amino acids like methionine, cystine, lysine and tryptophan are lacking in different sources of plant protein (Fetuga *et al.*, 1973; Felker and Bandurshi, 1977). But since animal protein sources are relatively more expensive, protein sources of plant origin have also been incorporated crustacean feeds by many workers to minimize the cost of production. Balazs *et al.* (1973) reported that the diets with soyabean meal gave results superior to that of fish-soyabean –shrimp based diet for *Macrobrachium rosenbergii.* Koshio *et al.* (1992) observed higher nutritive value of soyabean protein for juvenile *Macrobrachium rosenbergii* in comparison to crab protein concentrate. Tidwell *et al.* (1993) reported that fish meal can be partially or totally replaced with soyabean meal and distillers by-products in the diets for fresh water prawn, *Macrobrachium rosrenbergii.*

Moore and Stanley (1982) suggested that corn silage can play a role in supporting pond production of *Macrobrachium rosenbergii.* Pressed brewer's grain (by-products contained malted barley, corn, rice grit and hops) was evaluated as potential protein source for *Macrobrachium rosenbergii* (Kohler and Kruger, 1985). Ashmore *et al.* (1986) evaluated the nutritive merits of four cereal grains (corn, milo, wheat and barley) in formulated diets and obtained higher growth in *Macrobrachium rosenbergii* fed on barley based diet. Tidwell *et al.* (1992) reported that up to 40% of distillers dried grains with solubles can be used as a protein source in practical feeds of *Macrobrachium rosenbergii.*

Hari and Kurup (2003) found that there existed no significant difference in specific growth rate, protein efficiency ratio, weight gain and survival rate among juvenile of *Macrobrachium rosenbergii* fed varying plant –animal protein ratio. It would be possible to replace animal protein by low cost plant protein in prawn feed. Better growth performance in juveniles of *Macrobrachium rosenbergii* can be achieved by the incorporation of equal proportions of plant and animal protein (A: P=1) in the diet.

The effect of adding plant leaves including mangrove leaves and other plant materials in the diets for prawns have also been studied. AQUACOP (1976) reported that acacia meal or copra meal can be used as substitute ingredients in the diet of *Macrobrachium rosenbergii*, as the mineral and carotenoid requirements of the prawn may be met by these ingredients. Addition of fresh leaves (*Ailanthus altissima* and *Malva parviflora*) to be the diet of the prawn *Macrobrachium rosenbergii* resulted in elimination of Black Death Syndrome, and reduction in incidence of black spot (Harpaz and Schmalback, 1986). Graces and Heinen (1989) have shown that orange flesh, peeled sweet potatoes, frozen peeled bananas, turnip greens and carrot tops to be useful supplements with commercial trout chow for feeding prawns. Jayalakshmy and Natarajan (1993) reported that among four pelleted feeds prepared for *Macrobrachium idella* using *Pandina, Elodea, Lemna and Bruguiera*, *Elodea* proved best. Vasudevappa *et al.*(1993) obtained best growth in *Macrobrachium rosenbergii* fed on fish meal based diet incorporated with ground nut leaf powder than the diets either with fish meal alone or incorporated with mulberry leaf powder. Jeyalakshmi *et al.* (1997) obtained highest growth percentage in *Macrobrachium malcomsonii* fed on diet incorporated with leaf protein concentrate extracted from *Cajanus cayam* followed by *Chioria ternala*. Eusebio and Coloso (1998) evaluated the potential of locally available legumes (white cowpea,vigna unguiculata and green mungbean ,vigna radiate) and leaf meals (papaya,caricapapaya and cassava,manihut esculenta) in combination with defatted soyabean meal as a protein sources in juvenile *Penaeus indicus*. The shrimp fed with papaya and cassava leaf meals gave higher gain and SGR, which did not differ from those shrimp fed with cowpea meal.

2.2.3. Aquatic weeds

A number of aquatic weeds has been used in fish feeds replacing conventional ingredients(Ray and Das,1993; Routray and Routray,1997; Noor *et al.*,2000; Falaye *et al.*,2001; Fasakin *et al.*,2001; Bairagi *et al.*,2002; Landesman *et al.*,2002; Reyes and Fermin,2003). Duck weed, *Lemna minor* was used along with cabbage waste in the diet of common carp (Devaraj *et al.*, 1981) and *Lemna paucicostata* in the diets of Tilapia (Mbagwu *et al.*, 1989). Indian major carps (catla, rohu and mrigal) showed better FCR when, leaf meal

of two aquatic weeds (*Ottelia* and *Nymphoides*) were used as a protein source to replace fish meal in the supplementary feed (Patnaik et al., 1991). Inclusion of *pistia* at upto 45% level of inclusion in pelleted diets elicited good growth and food conversion in rohu (Ray and Das, 1995).

Two species of *Azolla* (*A. caroliniano and A. pinnata*) were used in the diets of carps at 60% and 80% inclusion levels respectively, replacing the conventional feed ingredients (Das *et al.*, 1994; Mohanty and Das, 1995). *Azolla* based diets when fed to tilapia and grass carp led to improved FCR and increase in fish yield. Basudha and Viswanath (1997) also found that, growth, FCR and PER of feed containing *Azolla* powder, fish meal, mineral and vitamin supplement was greatest compared to the feed containing rice bran and mustard oil cake. Haniffa *et al.* (2002) reported the incorporation of *Hydrilla verticellata* and drum stick leaves in the diet of *Macrobrachium idea*. Islam *et al.* (2004) safely used waste water grown duck weed as fish feed. Duck weed is better suited for feeding grass carp than pond weed or coontail.

The nutritional value and the possibility of utilization of the aquatic weed, *Nymphoides cristatum* as feed for young *Labeo rohita*, were evaluated in terms of growth, conversion efficiency, and specific activity of digestive enzymes, biochemical composition of flesh and microbial composition of intestine, in a 60 day laboratory feeding trial. The fish fed with the weed showed better average daily gain, FCR and PER. The specific activity of proteolytic and amylolytic enzymes showed significant increase in fish fed the weed. This study indicated the possibility of incorporation of the aquatic weed *N.cristatum* in the diet of *Labeo rohita* substituting conventional feed ingredients (Patra *et al.*, 2002).

2.2.4. Other sources

Single cell protein (SCP) included algal powders, yeast and bacterial protein. James *et al.* (1990) suggested that *Spirulina* cannot serve as a sole protein for post larvae of

Macrobrachium rosenbergii, but can be effectively used as a supplementary protein. Akiyama *et al.* (1992) reported that the levels of yeast should not exceed 5% in feeds unless product used is palatable to shrimp.

Manju and Devendran (1997) reported that *Macrobrachium idella* fed on diets containing bacteria and Actinomycetes gave significant growth, better conversion efficiency and increased body protein compared to fish meal based diet, suggesting that microbial single cell protein can replace the fish meal to a certain extent in formulated diets.

MATERIALS AND METHODS

3. MATERIALS AND METHODS

The experiment was done to work out the feasibility of using water spinach leaves as a low cost plant protein source in the feed of *Macrobrachium rosenbergii* post larvae and dietary evaluation of the formulated diet through growth trials and hence to determine the optimum level of substitution. *Ipomoea aquatica* Forsk, (Plate1) commonly known as water spinach is a dicotyledenous angiospermic plant belonging to the family Convolvulaceae. It is a perennial, trailing herb on wet muds but a floating plant in free waters, stem hollow, slender with white, spongy floats and rooting at the nodes. Leaves are elliptic or ovate-oblong. Flowers are white or pink in colour and bell shaped. Propagation is by seeds and stem fragments. Plant is common in India, south-east Asia, Taiwan and South China. The aquatic plant is cultivated. Its fresh young leaves and stems are used as vegetable, boiled or cooked in oil. In Malaysia water spinach has been grown and used as animal feed (Majid, 1998). The experiment was conducted at the Freshwater prawn hatchery, College of Fisheries, Panangad, Cochin over a period of 60 days.

3.1. PREPARATION OF THE FEED

3.1.1. Feed ingredients

Five experimental diets (T1, T2, T3, T4 and T5) were prepared for *Macrobrachium rosenbergii* post larvae. Control diet (T1) was prepared by using clam meat, wheat bran, ground nut oil cake, tapioca flour, sunflower oil and vitamin-mineral mixture. Different test diets (T2-T5) were prepared by replacing clam meat with *Ipomoea* leaf meal. The ingredient proportions of the experimental diets are given in the Table 1. The composition of Supplevit-M, the vitamin mineral mix incorporated in the feed is presented in Table2.

Plate 1. ***Ipomoea aquatica***

Table 1. Ingredient composition (g/100g) of experimental diets fed to *Macrobrachium rosenbergii* post larvae

Ingredients (%)	FEEDS				
	T1 (0% I)	T2 (12% I)	T3 (24% I)	T4 (36% I)	T5 (48% I)
Water spinach	-----	12	24	36	48
Clam meat	40	36	32	28	24
Wheat bran	40	32	23	11	7
GOC	10	10.25	11	12.5	11.5
Tapioca flour	8	7.75	8	10.5	7.5
Vitamin mineral mix	1	1	1	1	1
Sunflower oil	1	1	1	1	1
TOTAL	100	100	100	100	100

I-*Ipomoea* leaf meal

Table 2. Composition of* Supplevite-M (Vitamin mineral concentrate).

Each 250 mg provides	Quantity
Vitamin A	500,000 IU
Vitamin D3	100,000 IU
Vitamin B2	0.2g
Vitamin E	75 units
Vitamin K	0.1g
Calcium pantothenate	0.25g
Nicotinamide	1g
VitaminB12	0.6 g
Choline chloride	15g
Calcium	75g
Manganese	2.75g
Iodine	0.1g
Iron	0.75 g
Zinc	1.5g
Copper	0.2g
Cobalt	0.045g

***Supplevite-M: Sarabhai Chemicals, Mumbai**

3.1.2. Processing of the feed ingredients

Various ingredients used for feed formulation were processed as follows:

All the ingredients for formulation of the feeds were procured locally. *Ipomoea* leaves collected from water areas adjacent to the college were dried and powdered. Meat of the black clam *Villorita cyprinoids* was also washed, dried and powdered. The other ingredients namely wheat, bran, oil cake and tapioca flour were dried, powdered and sieved through 250μ mesh. The powdered ingredients were packed separately in air tight plastic bottles and kept in a refrigerator along with other ingredients till they were used for feed preparation.

3.1.3. Proximate composition of feed ingredients

Proximate composition of all the feed ingredients was analyzed prior to feed formulation. The methods used for the analysis are shown below:

Moisture content %: Drying the sample at 105°C till a constant weight was arrived

Ash content %: Ashing the sample at 100±10°C for 6 hours in a muffle furnace.

Crude fat %: Solvent extraction using petroleum ether (B. P 40- 60°C) in a soxhlet extraction apparatus for 6 hours.

Crude protein%: Microkjeldhal method (AOAC, 1990).

Crude fiber%: Pearson method (1976).

Nitrogen free extract %: 100-(%protein+%lipid+%ash+%fiber+%moisture)

By difference method (Hasting, 1976).

3.1.4. Formulation and processing of experimental diets.

Five types of pelleted feeds were formulated fixing their protein level at 30%.

The experimental feeds were prepared separately by mixing required quantity of ingredients. The respective ingredients were weighed accurately in an electronic balance and

all the ingredients except Supple-M and sunflower oil were mixed well with sufficient water to make a smooth dough. The dough was transferred to a vessel and steamed for 20 minutes in a pressure cooker. The steamed dough was cooled under fan. Sunflower oil and vitamin-mineral mixture were added to this and mixed well. Then it was extruded through a hand pelletizer and dried in an oven at 60°C to a moisture content of less than 12%. After drying, the pellets were broken into small pieces and packed in airtight plastic bottles.

3.1.5 Proximate composition of experimental diets

Proximate composition of the experimental diets was analyzed to evaluate the nutrient status. Methodology employed was the same as that of the ingredients for analysis.

3.2. EXPERIMENTAL ANIMALS

The post larvae of *Macrobrachium rosenbergii* derived from a single female hatch were procured from Prakruthi Aquatics, Perumbavoor and were transported to the College hatchery in oxygen filled polyethylene bags under minimum stress. Five hundred post larvae were introduced into an oval, flat bottom fiber glass tank of 1.2 ton capacity filled with filtered fresh water up to half the capacity and provided with gentle aeration. The post larvae were fed *ad libitum* with granulated artificial feed having clam meat as the chief source of protein. Leftover feed and waste were removed daily by siphoning out and 75% of water was renewed every day. Tiles and PVC pipes were provided at the bottom of the tank in order to reduce cannibalism. Twenty-five days old post larvae of uniform size collected from this were used for the present experiment. Ten post larvae were randomly distributed to all tanks after recording their initial weight.

3.3 EXPERIMENTAL SET UP

The experiment was carried out in the *Macrobrachium* hatchery of College of Fisheries, Panangad, roofed with translucent fiberglass reinforced plastic (FRP) sheets for moderate light conditions. Circular, flat bottom fiber glass tanks with the following specifications were used for the experiments.

Capacity of the tank-83litres
Diameter-55cm
Height-35cm
Rim width-3cm
Thickness of wall-4mm
Colour-Aquamarine

Clear well water filtered through a close meshed nylon blotting silk was used for filling the tanks up to a height of 25cm. Mild uniform aeration was provided in the tanks with air diffusion stones and control valves. Small PVC pipes were kept at the bottom of the tank in a slanting position for providing shelter to the animals.

3.4 EXPERIMENTAL DESIGN AND PROCEDURE

Flat bottomed circular fiber glass tanks were used for the experiment. Two hundred numbers of healthy uniform sized post larvae having an average weight of 0.077 g were selected from a population of 500 numbers and 10 numbers each randomly distributed in twenty experimental tanks. The experiment was conducted in a Completely Randomized Design with 5 treatments and 4 replications each. Before the commencement of feeding with the experimental diet, the post larvae were conditioned with the control diet for 5 days and they were acclimatized to tray feeding. Then 10 prawns from each tank were collected and weighed together in an electronic balance with a precision of 0.001g. Initial average weight of the post larvae was 0.077g.

Each treatment group of animals was fed with corresponding diets at the rate of 15% biomass for the first 30 days. After that, the feeding ration was reduced to 10% and maintained so till the end of the experiment. Petri dishes were kept at the bottom of the tank close to the substratum provided. Every day before offering the feed, leftover feed and faecal matter were collected and dried at 60 °C for estimation of FCR and crude digestibility. Petri dishes were cleaned thoroughly before next feeding.

The tanks were cleared off left - over feed and faecal matter daily before feeding. Water quality was maintained by regular replenishment as required.

Sampling was done at fortnightly intervals. At each sampling all the prawns stocked were taken and weighed. The quantum of feed given was adjusted based on the increased weight.

Carcass composition of the prawns were analysed initially and also on termination of the feeding trial following standard methods, the same as that for analysis of the ingredients.

3.5. WATER QUALITY PARAMETERS

During the experiment water quality parameters such as temperature, pH, Dissolved oxygen, ammonia and alkalinity were checked by the following methods.

Temperature: By using mercury thermometer of 0.1°C

pH: By using universal indicator solution

Dissolved oxygen: Standard Winkler method (Strickland and Parsons, 1972)

Ammonia: By Hypochlorite spectrophotometer method

Alkalinity: Tritrimetric method

Temperature and pH were measured daily and dissolved oxygen, ammonia and alkalinity once in a week.

3.6. CARCASS COMPOSITION

Before and after the experiment the prawns were subjected to proximate analysis for estimating body composition by the methods already described.

3.7. EVALUATION INDICES

The parameters evaluated are growth(Net weight gain), Specific growth rate (SGR), Percentage survival, Food Conversion Ratio (FCR) ,Food Conversion Efficiency (FCE),Protein efficiency ratio (PER),Crude protein digestibility coefficient and proximate composition of test animals.

3.7.1. Net weight gain

It gives the increase in the weight of prawns during the experimental period. It was calculated by using the formula.

Net weight gain= Final weight –Initial weight

3.7.2. Specific growth rate

Growth performance can be measured in terms of specific growth rate (SGR) since it is a more refined and improved growth index than absolute weight gain or percentage growth rate (Hepher, 1988). In the present study SGR was calculated by using the following formula.

$$SGR=\frac{\log w_2-\log w_1}{t2-t1}$$

Where, w_1=weight at day t_1

W_2=weight at day t_2

t_2-t_1= Duration of experiment in days.

3.7.3. Survival rate

It is expressed in terms of percentage

$$\text{Survival percentage}=\frac{\text{Initial number- number of dead prawn}}{\text{Initial number}}\times 100$$

3.7.4. Food conversion ratio

Food conversion ratio is the ratio between the weight of food consumed and the weight gain of the animal which often serves as a measure of efficiency of the diet.

$$FCR = \frac{\text{Average weight of food consumed in dry weight}}{\text{Average live weight gain}}$$

3.7.5. Food Conversion Efficiency (FCE)

Food conversion efficiency is the wet gain of animal from one unit of food consumed and this was calculated using the following formula.

$$\% \text{ FCE} = \frac{\text{Wet weight gain of prawn (g) during the sampling period X100}}{\text{Dry weight of food consumed (g) during the sampling period}}$$

3.7.6. Protein efficiency ratio

Protein efficiency ratio is the ratio between the weight gain of prawn and amount of protein consumed. It was calculated by employing the formula of Hepher (1988).

$$PER = \frac{\text{Wet weight gain of prawn (g)}}{\text{Crude protein consumed (g)}}$$

3.7.7. Crude protein digestibility coefficient

Protein digestibility was expressed as apparent digestibility of protein, calculated employing the following formula:

$$\text{Digestibility (\%)} = \frac{\text{Qty of protein consumed-Qty of protein in the faeces} \times 100}{\text{Quantity of protein consumed}}$$

3.8. STATISTICAL ANALYSIS

The experiment was carried out by using the Completely Randomized Design (CRD). All the evaluation indices were analyzed by using Analysis of variance (ANOVA) at 5% level significance. Pair wise comparisons of treatments were done using least significant difference.

RESULTS

4. RESULTS

The efficacy of *Ipomoea* leaf meal as a partial substituent for clam meat in diets for Macrobrachium *rosenbergii* post larvae was evaluated over a period of 60 days. The results of the experiment are presented below.

4.1. PROXIMATE COMPOSITION OF THE FEED AND THE FORMULATED PELLETED FEED

4.1.1. Feed ingredients

The data pertaining to proximate composition of the various ingredients used in formulating the pelleted feeds are given in Table 3.

The moisture content of the feed ingredients viz., *Ipomoea aquatica* leaf meal, clam meat, wheat bran, ground nut oil cake and tapioca flour ranged from 8.1 to12 %, with the maximum in *Ipomoea* leaf meal (12%) and the minimum in clam meat (8.1%). Clam meat had the highest percentage of crude protein (50.7%), followed by ground nut oil cake (42 %), *Ipomoea aquatica* leaf meal (24.9 %) and wheat bran (13.5 %). Tapioca flour had the minimum crude protein content of 1.8 %.

Table 3. Proximate composition of the ingredients used in the formulation of feeds.

Ingredients	moisture %	Crude protein%	crude fat %	ash %	Crude fibre%	Nitrogen free extract %
Ipomoea aquatica leaf meal	12	24.9	3.4	13	10.5	36.2
Clam meat	8.1	50.7	8.9	6.4	3.9	22.0
Wheat bran	10	13.5	2.6	3.0	12.2	58.7
Ground nut oil cake(GOC)	10	42	7.3	2.5	13.0	25.2
Tapioca flour	11.5	1.8	1.3	2.3	2.0	81.1

4.1.2. Formulated pelleted feeds

The proximate composition of the formulated feeds is presented in Table 4. The percentage of moisture in the five feeds varied between 9 %(feedT3) and 11.8 % (feed T4), whereas crude protein content ranged from 33.75 % (feedT2, T4 andT5) to 34.99 % (feed T1 and T3). The crude fat content ranged from between 4.2% (feedT1) to 5.2% (feed T2). The ash content ranged from 5% (feedT1) to 7% (feed T3).

Table 4. Proximate composition of test diets used in the experiment

Parameters (%)	FEEDS				
	T1	T2	T3	T4	T5
moisture	10.4	11.4	9	11.8	10.2
Crude protein	34.99	33.75	34.99	33.75	33.75
Crude fat	4.2	5.2	5.08	5.09	4.98
Ash	5.0	5.8	7.0	6.7	6.9
Fibre	0.49	0.56	0.65	0.91	1.25
Nitrogen free extract	44.92	43.29	43.28	41.75	42.92

4.1.3. Water stability of feed

Test for water stability of different test diets was done for varying periods such as 1 hour, 2 hours, 4 hours and 6 hours. The results are given in Table 5. Maximum water stability was for the feed T3 and minimum for T5.

Table 5. Percentage water stability of test diets

feed	1hr	2hr	4hr	6hr
T1	95.64	93.23	90.05	88.46
T2	94.25	92.63	89.16	88.56
T3	95.00	93.44	91.61	89.61
T4	94.07	93.06	82.80	85.22
T5	93.62	90.01	80.83	82.00

4.2. EFFICIENCY OF TEST DIETS

4.2.1. Growth

Data regarding average live weight gain of prawns fed on the experimental feeds are presented in Table 6.

The average live weight gain of *Macrobrachium rosenbergii* post larvae fed on different experimental diets T1,T2,T3,T4 and T5 was found to be 0.3594g,0.6036 g0.,5687 g,0.3127 g and 0.3002 g respectively. The highest average live weight gain of 0.6036 g was obtained in treatment T2 with 12 % *Ipomoea* and lowest average weight gain of 0.3002g was in treatment T5 with 48% *Ipomoea.*

Analysis of variance showed that the growth of prawn fed on different feeds with varying levels of ipomoea leaf meal was significantly different ($p<0.05$). Pair-wise comparison showed that T4 and T5 were at par and the average gain in weight was lower than that of control (T1) for both the treatments. Effect of treatments T1, T2, T3 were entirely different ($p<0.05$). But the average gain in weight was the highest for T2 followed by T3 and T1.

The average percentage weight gain was found to be 474.61, 809.63, 751.2, 419.20 and 400.18 respectively for T1, T2, T3, T4 and T5. The highest percentage gain was found to be obtained in treatment T2 with 12% *Ipomoea* and lowest in treatment T5 with 48% *Ipomoea.* Graphical representation of net weight gain for different treatments is shown in fig- 1

Table 6. Growth of *Macrobrachium rosenbergii* post larvae fed on experimental diets.

Treatment	Replication	Average initial wt (g)	Average final wt(g)	Av wt gain (g)	Mean ±SD	% wt gain	Mean±SD
T1	1	0.0764	0.3886	0.3122	0.3594 ±1.079	408.6	474.6125 ±38.9353
	2	0.0783	0.4596	0.3813		486.9	
	3	0.0773	o.4708	0.3935		509.05	
	4	0.0710	0.4217	0.3507		493.9	
T2	1	0.0733	0.6743	0.6010	0.6036± 1.811	819.9	809.625 ±33.9175
	2	0.0759	0.6741	0.5982		788.1	
	3	0.0725	0.6959	0.6234		859.8	
	4	0.0768	0.6687	0.5919		770.7	
T3	1	0.0762	0.6426	0.5664	0.5687± 1.706	743.3	751.195 ±15.5388
	2	0.0733	0.6152	0.5419		739.2	
	3	0.0758	0.6655	0.5897		777.9	
	4	0.0775	0.6544	0.5769		744.38	
T4	1	0.0740	0.3805	0.3065	0.3127± 0.938	414.18	419.1975 ±13.5466
	2	0.0757	0.3871	0.3114		411.36	
	3	0.0748	0.3806	0.3058		408.82	
	4	0.0740	0.4014	0.3274		442.43	
T5	1	0.0725	0.3606	0.2881	0.30027± 0.987	397.37	400.0175 ±8.1781
	2	0.0787	0.3866	0.3079		391.2	
	3	0.0739	0.3681	0.2942		398.1	
	4	0.0752	0.3861	0.3109		413.4	

Table 7. Analysis of variance of growth of *Macrobrachium rosenbergii* fed on experimental diets.

Source of variation	Sum of squares	Degrees of freedom	Mean sum of squares	F Value
Treatment	0.33974	4	0.08493	198.9899*
Error	0.006403	15	0.000427	
Total	0.346147	19		

*Significant at 5%level

Comparison of treatment means based on critical difference:

Critical difference=0.03114

Treatment	T2	T3	T1	T4	T5
Mean	0.60363	0.568725	0.35943	0.31278	0.30028

Underscored means are not significantly different

4.2.2. Specific growth rate

The data on the specific growth rate of prawns under various treatments are given in Table 8.

Analysis of variance of the data showed significant difference ($p<0.05$) in SGR values between treatments (Table 9).

Table 8.Specific growth rate of *Macrobrachium rosenbergii* fed on experimental diets.

Treatment	Replication	Av initial wt (g)	Av final wt(g)	Av wt gain (g)	Specific growth rate	Mean ±SD
T1	1	0.0764	0.3886	0.3122	2.71	2.91±0.1155
	2	0.0783	0.4596	0.3813	2.94	
	3	0.0773	o.4708	0.3935	3.01	
	4	0.0710	0.4217	0.3507	2.96	
T2	1	0.0733	0.6743	0.6010	3.69	3.67±0.06124
	2	0.0759	0.6741	0.5982	3.63	
	3	0.0725	0.6959	0.6234	3.76	
	4	0.0768	0.6687	0.5919	3.60	
T3	1	0.0762	0.6426	0.5664	3.55	3.57±0.0324
	2	0.0733	0.6152	0.5419	3.54	
	3	0.0758	0.6655	0.5897	3.62	
	4	0.0775	0.6544	0.5769	3.55	
T4	1	0.0740	0.3805	0.3065	2.72	2.74±0.0421
	2	0.0757	0.3871	0.3114	2.71	
	3	0.0748	0.3806	0.3058	2.71	
	4	0.0740	0.4014	0.3274	2.81	
T5	1	0.0725	0.3606	0.2881	2.67	2.68±0.02598
	2	0.0787	0.3866	0.3079	2.65	
	3	0.0739	0.3681	0.2942	2.67	
	4	0.0752	0.3861	0.3109	2.72	

Table9. Analysis of variance of the specific growth rates of *Macrobrachium rosenbergii* fed on experimental diets.

Source of variation	sum of squares	degrees of freedom	mean sum of squares	F value
Treatment	3.45	4	0.8625	207.33*
Error	0.0624	15	0.00416	
Total	3.5124	19		

* Significant at 5% level

Comparison of treatment means based on critical difference:
Critical difference=0.0972

Treatments	T2	T3	T1	T4	T5
Mean	3.67	3.57	2.91	2.74	2.68

Underscored means are not significantly different

The prawns in treatment T2 gave highest SGR (3.67), followed by those in treatment T3 (3.57), control T1 (2.91), T4 (2.74) and T5 (2.68). The SGR of prawns in the treatments T1 (0% Ipomoea), T2 (12% Ipomoea), and T3 (24% Ipomoea) were significantly different and treatments T2 and T3 gave higher SGR values than that of T1. T4 (36% Ipomoea) and T5 (48% Ipomoea) belongs to the same homogeneous group and the SGR values were lower than that of control T1.

The SGR values of prawns fed on feeds containing different experimental diets are graphically represented in Fig - 2.

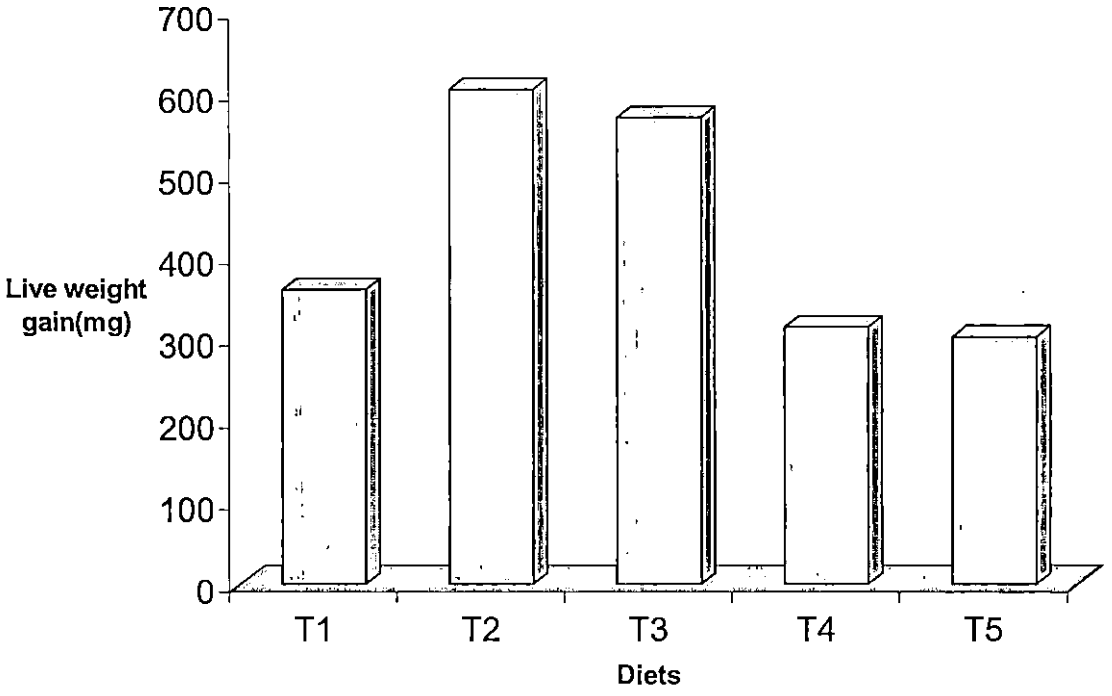

Fig.1.Growth of *M.rosenbergii* post larvae fed on the experimental diets

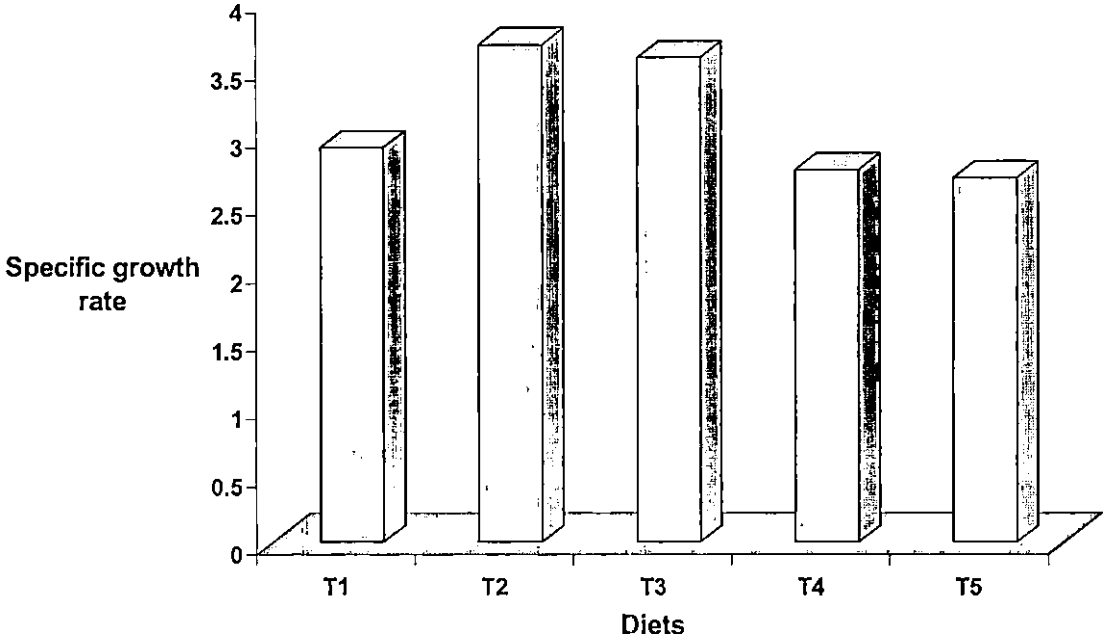

Fig.2. Specific growth rate of *M.rosenbergii* post larvae fed on the experimental diets

4.2.3. Survival

The data on survival per cent of *M. rosenbergii* post larvae fed on different experimental diets are given in Table 10.

The analysis of variance (Table 11) of the data on survival percentage showed no significant difference between the treatments.

Highest average survival percentage of 82.5% was obtained in treatmentT3 (24% Ipomoea), equivalent to control T1(without Ipomoea) followed by survival rates of 80% in treatment T2(12% Ipomoea) and T4(36 % Ipomoea) and 77.5% in treatment T5(48 % Ipomoea) respectively. These five treatments have insignificant influence on survival rate.

Graphical representation of percentage survival rates for different treatments are shown in Fig -3

Table 10. Percentage survival of *M.rosenbergii* post larvae fed on different experimental diets

Treatment	Replication	Initial No:	Final No:	%survival	Mean± SD
T1	1	10	7	70	82.5±10.8972
	2	10	10	100	
	3	10	8	80	
	4	10	8	80	
T2	1	10	7	70	80±15.8113
	2	10	10	100	
	3	10	8	80	
	4	10	7	70	
T3	1	10	10	100	82.5±17.85357
	2	10	10	100	
	3	10	7	70	
	4	10	6	60	
T4	1	10	6	60	80±12.2475
	2	10	8	80	
	3	10	9	90	
	4	10	9	90	
T5	1	10	9	90	77.5±10.8972
	2	10	8	80	
	3	10	6	60	
	4	10	8	80	

Table 11. Analysis of variance of survival rate of *M.rosenbergii* fed on different experimental diets

Source of variation	Sum of squares	Degrees of freedom	Mean sum of squares	F Value
Treatment	70	4	17.5	0.076
Error	3425	15	228.33	
Total	3495	19		

(F value is not significant at 5% level)

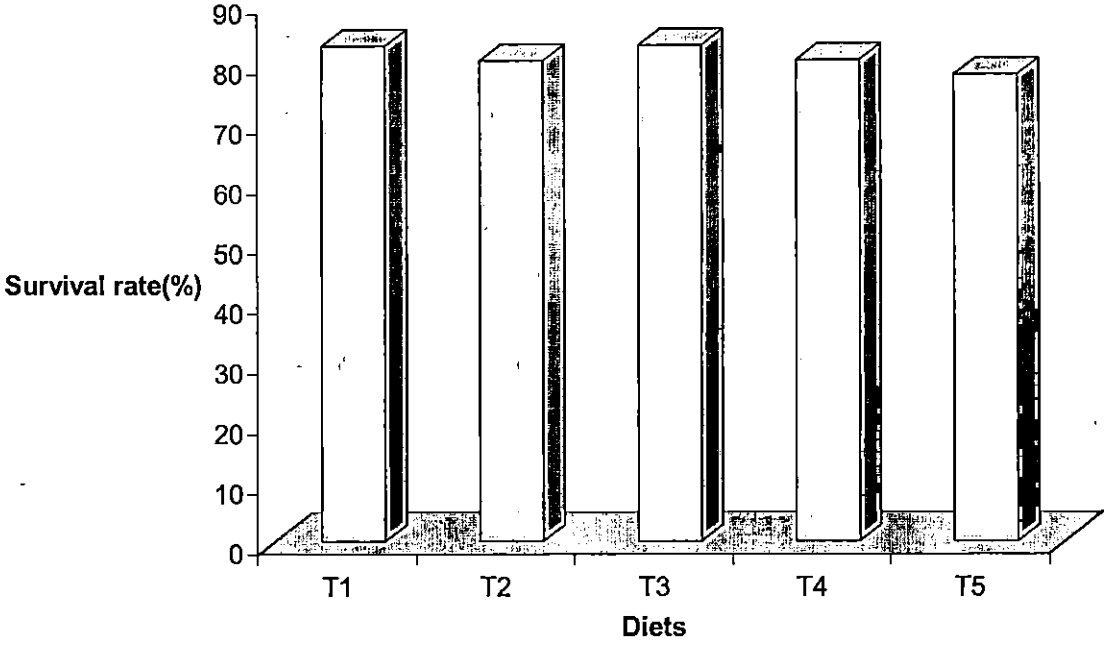

Fig.3. Percentage survival of *M.rosenbergii* post larvae fed on the experimental diets

4.2.4. Food conversion ratio

Food conversion ratio values of *M.rosenbergii* post larvae fed on different experimental diets are given in Table 12. The mean FCR values ranged from 1.11 to 2.275 in various treatments. The lowest FCR was obtained in treatment T2 (1.11) followed by T3 (1.2), T1 (1.9), T4 (2.125) and T5 (2.275).

Analysis of variance of FCR values showed significant difference ($p<0.05$) between various treatments (Table13). Pair wise comparison divided the treatments in to different groups. The feeds T2 (12% Ipomoea) and T3(24% Ipomoea) were effectively converted into body mass than that of control T1 (with out Ipomoea). The treatment T4 (36% Ipomoea) and T5 (48% Ipomoea) were not significantly different.

Graphical representation of FCR values of post larvae in various treatments are given in Fig-4

4.2.5. Food conversion efficiency

Food conversion efficiency values of *M. rosenbergii* post larvae fed on different experimental diets are given in Table 14. The mean FCE values ranged from 43.55 to 89.8 in various treatments.

The highest FCE was obtained in treatment T2 (089.8), followed by T3 (0.83). The FCE of treatmentsT1, T4 and T5 were 0.525.0.468 and 0.435 respectively. Treatments T2, T3 and T1 were significantly different among themselves. It was also found that treatments T4 and T5 were alike. The feeds T4 and T5 were less efficient in feed conversion than that of all other treatments. Analysis of variance of FCE values shows significant difference ($p<0.05$) between various treatments (Table15).

Graphical representation of FCE values of post larval prawn in various treatments are given in Fig-5

Table 12. Food Conversion Ratio of *M.rosenbergii* post larvae fed on different experimental diets

Treatment	replication	Av initial wt (g)	Av final wt(g)	Av wt gain (g)	Av feed consumed	Food Conversion Ratio	Mean±SD
T1	1	0.0764	0.3886	0.3122	0.68684	2.2	1.9 ±0.17321
	2	0.0783	0.4596	0.3813	0.68634	1.8	
	3	0.0773	o.4708	0.3935	0.7083	1.8	
	4	0.0710	0.4217	0.3507	0.63126	1.8	
T2	1	0.0733	0.6743	0.6010	0.6611	1.1	1.11 ±o.05745
	2	0.0759	0.6741	0.5982	0.65802	1.1	
	3	0.0725	0.6959	0.6234	0.64834	1.04	
	4	0.0768	0.6687	0.5919	0.70908	1.2	
T3	1	0.0762	0.6426	0.5664	0.67968	1.2	1.2±0
	2	0.0733	0.6152	0.5419	0.65028	1.2	
	3	0.0758	0.6655	0.5897	0.70764	1.2	
	4	0.0775	0.6544	0.5769	0.69228	1.2	
T4	1	0.0740	0.3805	0.3065	0.64365	2.1	2.125 ±0.06591
	2	0.0757	0.3871	0.3114	0.68508	2.2	
	3	0.0748	0.3806	0.3058	0.67276	2.2	
	4	0.0740	0.4014	0.3274	0.6548	2.0	
T5	1	0.0725	0.3606	0.2881	0.66263	2.3	2.275 ±0.02469
	2	0.0787	0.3866	0.3079	0.70817	2.3	
	3	0.0739	0.3681	0.2942	0.67666	2.3	
	4	0.0752	0.3861	0.3109	0.68398	2.2	

Table 13. Analysis of variance of food conversion ratios of *M.rosenbergii* post larvae fed on different experimental diets.

Source of variation	Sum of squares	Degrees of freedom	Mean sum of squares	F value
Treatment	4.58772	4	1.14693	103.32*
Error	0.1666	15	0.0111	
Total	4.75432	19		

*Significant at 5% level

Comparison of treatment means based on critical difference:

Critical difference=0.1587

Treatments	T2	T3	T1	T4	T5
Mean	1.11	1.2	1.9	2.125	2.275

Underscored means are not significantly different

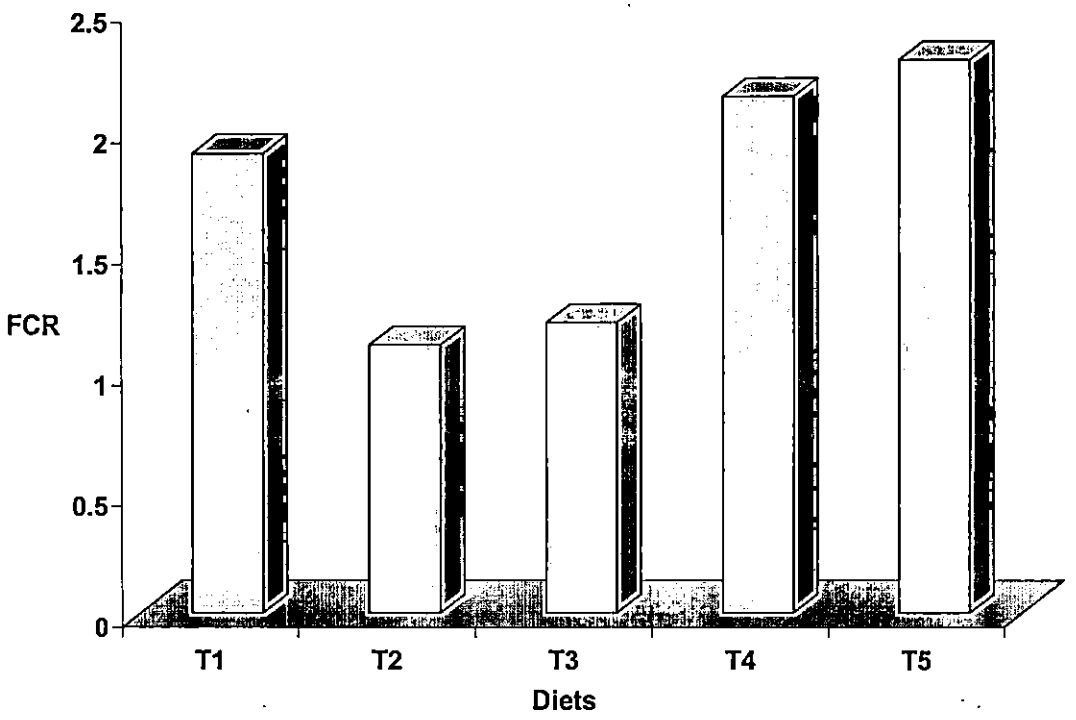

Fig.4. Food conversion ratio of *M.rosenbergii* post larvae fed on the experimental diets

Table 14. Food Conversion Efficiency of M.rosenbergii post larvae fed on different experimental diets.

Treatment	Replication	Food Conversion Efficiency (%)	Mean±SD
T1	1	45	52.5±0.0433
	2	55	
	3	55	
	4	55	
T2	1	90	89.75±0.05801
	2	90	
	3	96	
	4	83	
T3	1	83	83±0
	2	83	
	3	83	
	4	83	
T4	1	47	46.76±0.02046
	2	45	
	3	45	
	4	50	
T5	1	43	43.5±0.008660
	2	43	
	3	43	
	4	45	

Table 15. Analysis of variance of feed conversion efficiency of *M.rosenbergii* post larvae fed on different experimental diets.

Source of variation	Sum of squares	Degrees of freedom	Mean sum of squares	F Value
Treatment	0.74798	4	0.186995	155.83*
Error	0.018	15	0.0012	
Total	0.76598	19		

* Significant at 5%level

Comparison of treatment means based on critical difference:
Critical difference=0.05219

Treatments	T2	T3	T1	T4	T5
Mean	0.8975	0.83	0.525	0.4676	0.435

Underscored means are not significantly different

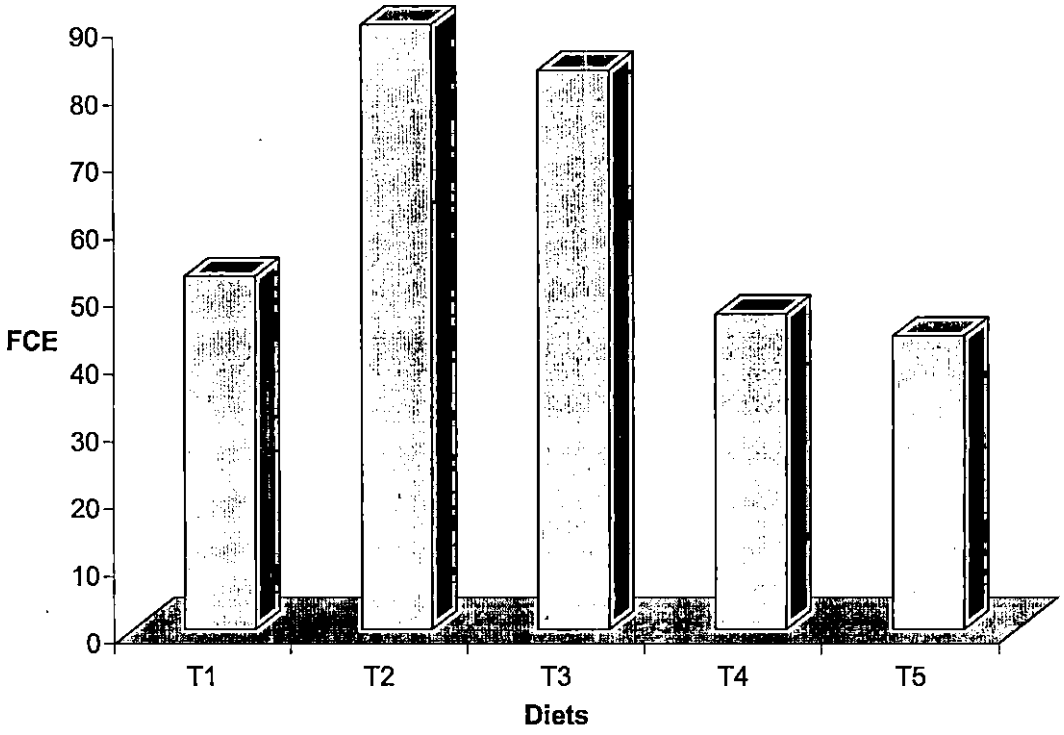

Fig.5. Food conversion efficiency of *M.rosenbergii* post larvae fed on the experimental diets

4.2.6. Protein efficiency ratio

The data on protein efficiency ratio of various treatments are given in Table 16.

The highest PER values was obtained in the treatment T2 (2.998) and the least in T5 (1.481). The average PER values obtained in various treatmentsT1, T3 and T4 were 1.756, 2.782 and 1.552 respectively. There was significant difference between treatments T2, T3 and T1. The treatments T4 and T5 were not significantly different. The PER values of T2 and T3 were higher than that of all other treatments.

Analysis of variance (Table17) of the data on PER values shows significant difference ($p<0.05$) among various treatments.

The PER values obtained in various treatments are graphically represented in Fig-6

Table 16. Protein Efficiency Ratio of *M.rosenbergii* post larvae fed on different experimental diets

Treatment	Replication	Av wt gain (g)	Crude protein (g)	Protein efficiency ratio	Mean ±SD
T1	1	0.3122	0.2068	1.51	1.7568±0.14551
	2	0.3813	0.2115	1.803	
	3	0.3935	0.2088	1.885	
	4	0.3507	0.1917	1.829	
T2	1	0.6010	o.1980	3.036	2.998±0.1255
	2	0.5982	0.2049	2.919	
	3	0.6234	0.1958	3.184	
	4	0.5919	0.2074	2.854	
T3	1	0.5664	0.2058	2.752	2.782±0.0578
	2	0.5419	0.1979	2.738	
	3	0.5897	0.2047	2.881	
	4	0.5769	0.2093	2.756	
T4	1	0.3065	0.1998	1.534	1.5522±0.0499
	2	0.3114	0.2045	1.523	
	3	0.3058	0.2020	1.514	
	4	0.3274	0.1999	1.638	
T5	1	0.2881	0.1959	1.471	1.481±0.0303
	2	0.3079	0.2125	1.449	
	3	0.2942	0.1996	1.474	
	4	0.3109	0.2031	1.531	

Table 17. Analysis of variance of protein efficiency ratio of *M.rosenbergii* fed on different experimental diets.

Source of variation	Sum of squares	Degrees of freedom	Mean sum of squares	F value
Treatment	8.286	4	2.0715	178.57*
Error	0.1753	15	0.0116	
Total	8.4613	19		

*Significant at 5%level

Comparison of treatment means based on critical difference:

Critical difference=0.1623

Treatment	T2	T3	T1	T4	T5
Mean	2.998	2.782	1.7568	1.5522	1.481

Underscored means are not significantly different

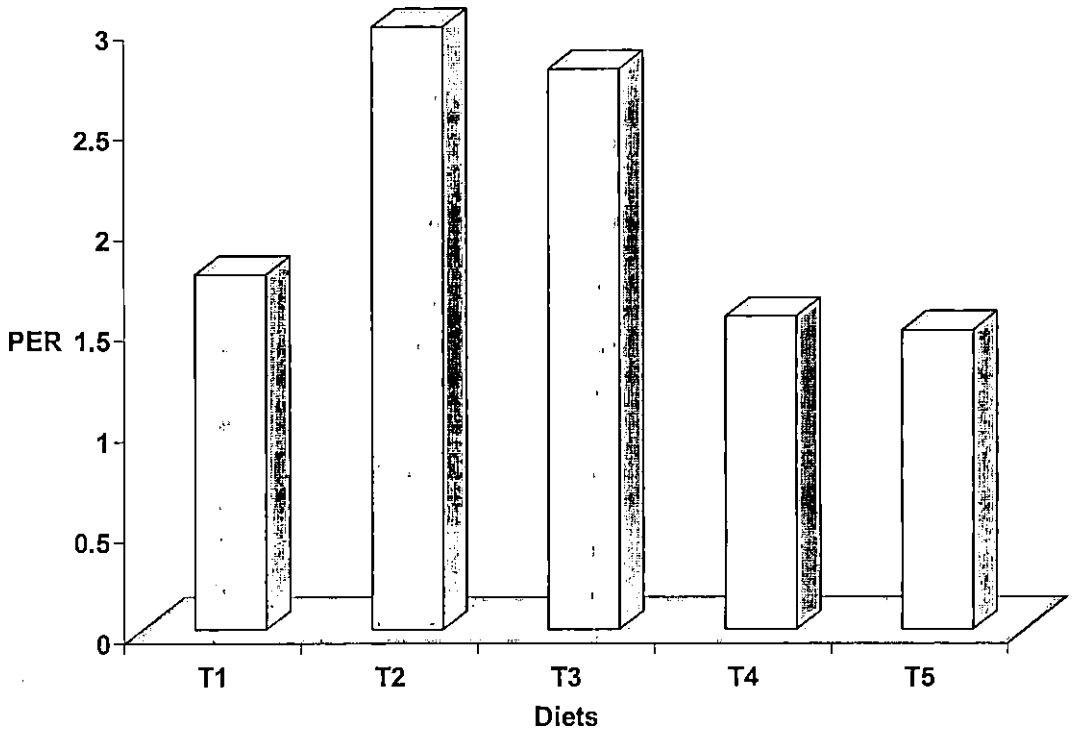

Fig.6. Protein efficiency ratio of *M.rosenbergii* fed on the experimental diets

4.2.7. Crude protein digestibility coefficient

The data on crude digestibility coefficient of various treatments are given in Table 18.

The highest crude digestibility coefficient was obtained in treatment T2 (84.12%) and least in T5 (68.42%). The average crude digestibility coefficient values obtained in other treatmentsT1, T3 and T4 were 74.02%, 82.39% and 74.41% respectively. The crude digestibility coefficient obtained in various treatments are graphically represented in Fig-7

Table 18.Crude protein digestibility coefficient of *M.rosenbergii* post larvae fed on different experimental diets

Treatments	protein consumed (g)	protein in faeces(g)	Digestibility coefficient (%)
T1	0.1193	0.031	74.02
T2	0.11333	0.018	84.12
T3	0.11922	0.021	82.39
T4	0.11333	0.029	74.41
T5	0.114	0.036	68.42

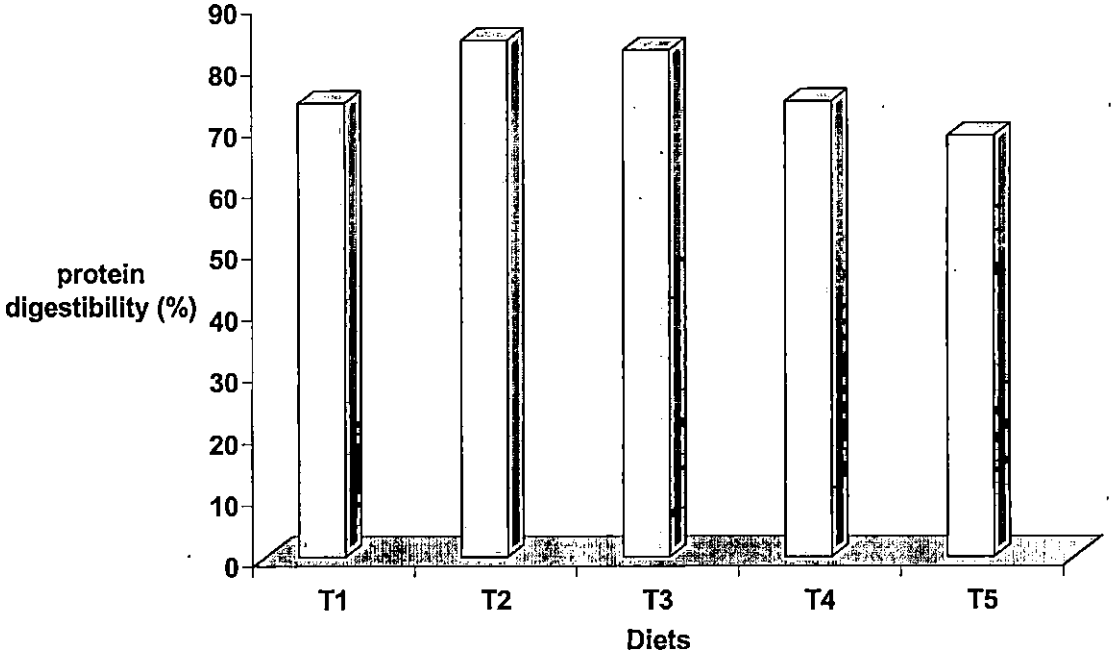

Fig.7. Protein digestibility coefficient of *M.rosenbergii* post larvae fed on experimental diets

4.3. Water quality parameters

4.3.1. Water temperature

The range of water temperature in the experimental tanks during the study period is given in Table 19. Minimum temperature recorded was 26°C and maximum temperature was 30.6°C. Weekly mean temperature values ranged from 26.1 to 30.3°C.

4.3.2. pH

Range of pH in the experimental tanks is given in the Table 20 .Minimum and maximum pH values observed during the study period were 6.9 and 8.4 respectively. Weekly mean pH values ranged from 7 to 8.2.

4.3.3. Dissolved oxygen

Range of dissolved oxygen (D.O) in the experimental tanks is given in Table 21. A minimum of 6.3 ppm, and a maximum DO content of 8.39ppm were obtained during the study period. Weekly mean values ranged from 6.67 to 7.8ppm.

4.3.4. Total alkalinity

Ranges of total alkalinity in the experimental tanks are given in table 22. A minimum of 90 ppm and a maximum of 127 ppm were obtained during the study period. Weekly mean values ranged from 99 to 112 ppm.

4.3.5. Ammonia

Ranges of ammonia in the experimental tanks is given in Table 23. A minimum of 0.056 mg/l and a maximum of 0.089 mg/l were obtained during the study period. Weekly mean values ranged from 0.0599 to 0.0716.

Table 19. Water temperature in the experimental tanks during the study period.

Temperature (°c)	Weeks							
	1	2	3	4	5	6	7	8
Mean	26.8	28	27.9	26.1	28.6	29	30.3	29.5
Range	26.7-26.9	27.6-28.4	27.7-28.1	26-26.2	28.3-28.9	28.8-29.2	30-30.6	29.3-29.7

Table 20. pH of water in the experimental tanks during the study period.

pH	Weeks							
	1	2	3	4	5	6	7	8
Mean	8.2	8	8.2	7.2	8	7	8.1	7.2
Range	8-8.4	7.9-8.1	8-8.3	7-7.4	7.8-8.2	6.9-7.1	8-8.3	7-7.4

Table 21. Dissolved oxygen of water in the experimental tanks during the study period

Dissolved oxygen (ppm)	Weeks							
	1	2	3	4	5	6	7	8
Mean	7.8	7.3	7.8	6.67	7.59	7.46	7.53	6.82
Range	7.5-7.8	7.1-8.1	7.5-8.1	6.3-7.01	7.08-8.28	6.62-8.39	7-8.16	6.34-7.25

Table 22. Total alkalinity of water in the experimental tanks during the study period

Total alkalinity (ppm)	WEEKS							
	1	2	3	4	5	6	7	8
Mean	112	105	110	103	106	99	106	104
Range	105-120	90-115	103-127	98-112	95-115	93-111	98-115	97-110

Table 23. Ammonia of water in the experimental tanks during the study period

NH3-N mg/l	WEEKS							
	1	2	3	4	5	6	7	8
Mean	0.0692	0.0658	0.0716	0.0599	0.068	0.0698	0.0678	0.067
Range	0.060-0.089	0.056-0.074	0.061-0.086	0.059-0.070	0.06-0.080	0.06-0.075	0.059-0.074	0.061-0.074

4.4. Carcass proximate composition

At the beginning and end of the experiment the prawns were subjected to biochemical analysis. Crude protein, crude fat, ash and carbohydrate are expressed as percentage of wet body weight.

4.4.1. Initial carcass proximate composition of test animals

The carcass compositions of the test animals were estimated and the corresponding data are given in Table 24. Moisture, protein, lipid, ash and carbohydrate levels were 80.02 %, 9.32 %, and 3.91 %, 1.09 % and 5.66 % respectively.

4.4.2. Final carcass proximate composition of test animals

Final carcass proximate composition of *M.rosenbergii* post larvae were evaluated after the experimental period of sixty days and the data are given in Table 25.

From the data it is clear that moisture of the body of test animals were maximum for the control diet (T1) and minimum for treatment T5. The moisture content of the diet T1 was significantly different from all other test diets containing *Ipomoea* leaf meal. Protein of the body of test animals was higher for the treatmentT3 and minimum for treatmentT1. The post larval prawn fed with *Ipomoea aquatica* leaf meal exhibited higher protein content ranging from 10.1 to 10.8 % on wet weight basis compared to control diet. Similarly the carbohydrate content was significantly higher in diets containing *Ipomoea* leaf meal (T2, T3, T4 and T5), ranged between 3.4 to 4.2 %, where as control diet (T1) had only 1.3 %.

Fat content was higher in control diet T1, where as *Ipomoea* based diets showed lower fat content (4.1 to 4.5%). The control diet T1 was significantly different from *Ipomoea* based test diets (T2-T5).

The ash content of the body of test animals was higher for T2 followed by T5 and T3. The treatments T2, T3 and T5 belonged to the same homogeneous group and T1 and T4 were significantly different. The maximum ash content in the test diet T2 was 1.04 and minimum in the test diet T1 (0.6%). Carbohydrate content of *M.rosenbergii* fed on different

experimental diets were found to be significantly different. Treatments T2 and T3 showed no significant difference.

The result of analysis of variance of moisture, crude protein, crude fat, ash and carbohydrate are given in Table26, 27, 28,29and 30 respectively. Graphical representation of percentage moisture, protein and fat content of body of the test animals are given in Fig-8, 9 and10.

Table 24. Initial carcass composition of post larvae of *M.rosenbergii*

Mean±S.D	moisture %	crude protein%	crude fat%	ash%	carbohydrate%
	80.017±0.4617	9.317±0.0021	3.91±0.0038	1.097±0.0256	5.659±0.0235

Table 25. Final carcass composition (mean ±SD)* *of M.rosenbergii* post larvae fed on the experimental diets

Parameter	T1	T2	T3	T4	T5
Moisture ± SD	84.5±0.7901	80.7±0.7036	80.36±0.7219	80.56±0.5694	79.75±0.60
Ash± SD	0.6±0.0707	1.043±0.0602	0.9375±0.0822	0.895±0.0503	0.950±0.06
Fat ± SD	5.45±0.1118	4.350±0.2062	4.40±0.0707	4.18±0.0831	4.500±0.07
Protein± SD	9.10±0.1225	10.175±0.0829	10.800±0.0707	10.425±0.0433	10.475±0.0
Carbohydrate± SD	1.314±0.4164	3.600±0.2774	3.407±0.2716	3.97±0.0923	4.23±0.242

*Average of three replications expressed as wet weight basis

Table 26. ANOVA of moisture content of *M.rosenbergii* fed on the experimental diets

Source of variation	Sum of squares	Degrees of freedom	Mean sum of squares	F value
Treatment	57.4337	4	14.35843	23.0458*
Error	9.3456	15	0.62304	
Total	66.7795	19		

* Significant at 5% level

Comparison of treatment means based on critical difference:

Critical difference=1.18939

Treatment	T1	T2	T4	T3	T5
Mean	84.5	80.7	80.56	80.36	79.75

Underscored means are not significantly different

Table 27. ANOVA of protein content of *M.rosenbergii* fed on the experimental diets

Sources of variation	Sum of squares	Degrees of freedom	Mean sum of squares	F value
Treatment	6.7875	4	1.6969	179.24*
Error	0.142	15	0.009467	
Total	6.9295	19		

*Significant at 5%level

Comparison of treatment means based on critical difference:

Critical difference=0.1466

Treatments	T3	T5	T4	T2	T1
Mean	10.8	10.475	10.425	10.175	9.1

Underscored means are not significantly different

Table 28. ANOVA of fat content of *M.rosenbergii* fed on the experimental diets

Sources of variation	Sum of squares	Degrees of freedom	Mean sum of squares	F value
Treatment	4.05	4	1.0125	53.01*
Error	0.287	15	0.0191	
Total	4.337	19		

*Significant at 5% level

Comparison of treatment means based on critical difference:

Critical difference=0.2083

Treatments	T1	T5	T3	T2	T4
Mean	5.45	4.5	4.4	4.35	4.18

Underscored means are not significantly different

Table 29. ANOVA of Ash content of *M.rosenbergii* fed on the experimental diets

Sources of variation	Sum of squares	Degrees of freedom	Mean sum of squares	F value
Treatment	0.4525	4	0.1131	19.9524*
Error	0.0847	15	0.00567	
Total	0.5373	19		

*Significant at 5% level

Comparison of treatment means based on critical difference:

Critical difference=0.11346

Treatments	T2	T5	T3	T4	T1
Mean	1.0425	0.95	0.9375	0.895	0.6

Underscored means are not significantly different

Table 30. ANOVA of carbohydrate content of *M.rosenbergii* fed on the experimental diets

sources of variation	Sum of squares	Degrees of freedom	Mean sum of squares	F value
Treatment	21.472	4	5.368	62.455*
Error	1.2893	15	0.08595	
Total	22.7613	19		

*Significant at5% level

Comparison of treatment means based on critical difference:

Critical difference=0.4418

Treatment	T5	T4	T3	T2	T1
Mean	4.234	3.9703	3.60012	3.4078	1.3139

Underscored means are not significantly different

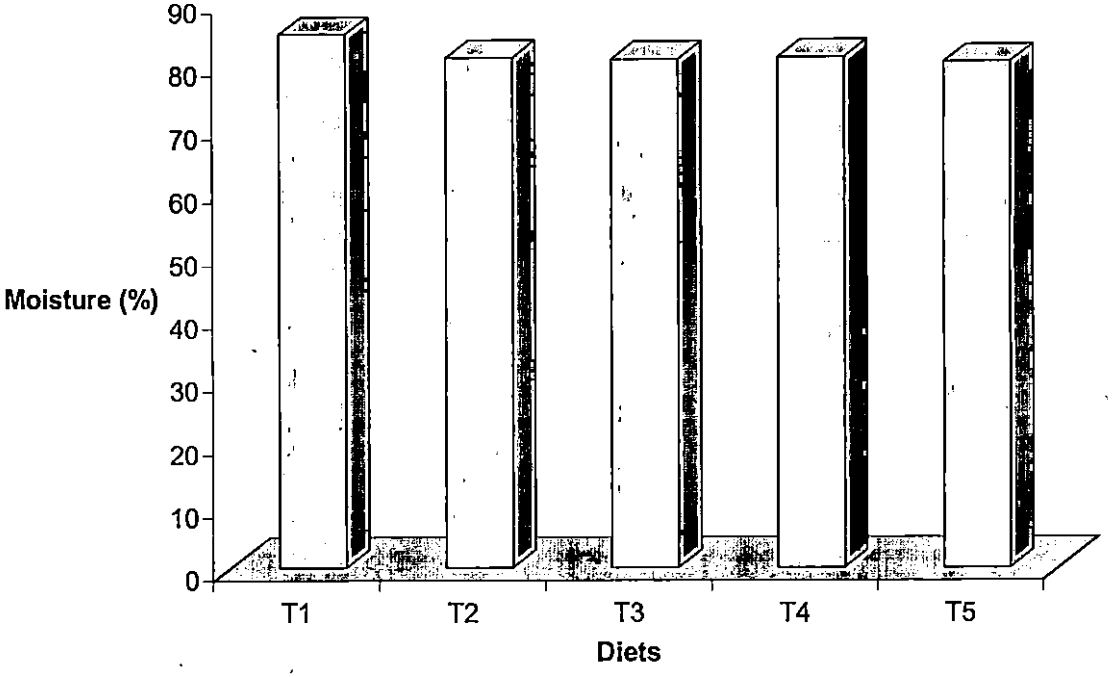

Fig.8.Final moisture content in the body of *M.rosenbergii* at the end of the experiment

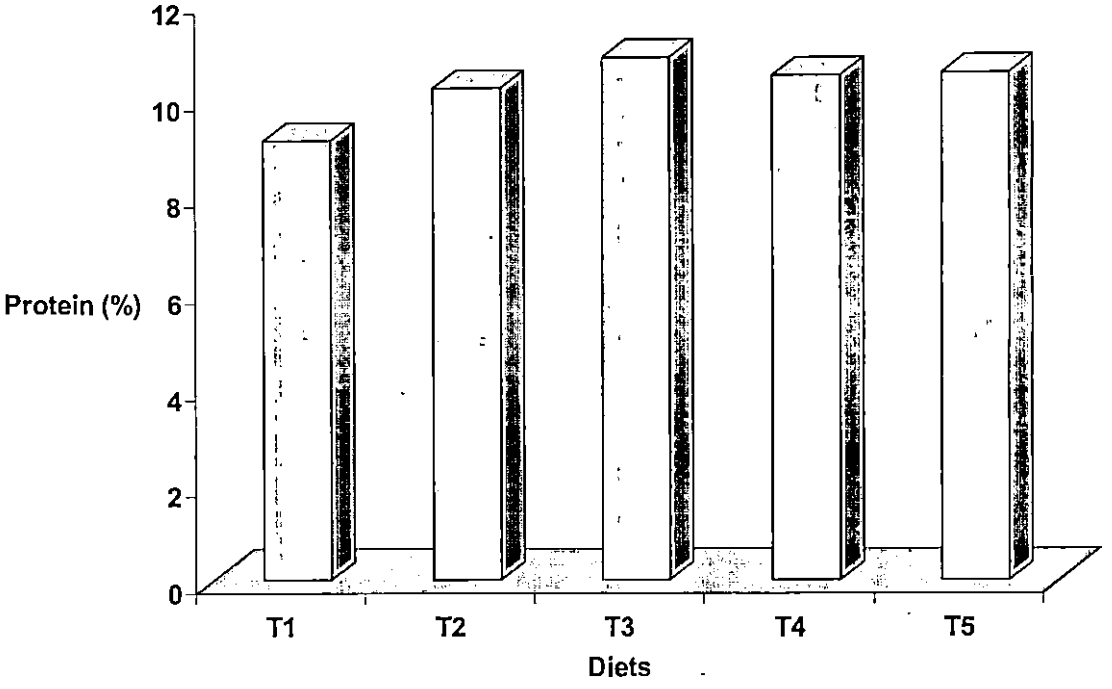

Fig.9.Final protein content in the body of *M.rosenbergii* at the end of the experiment

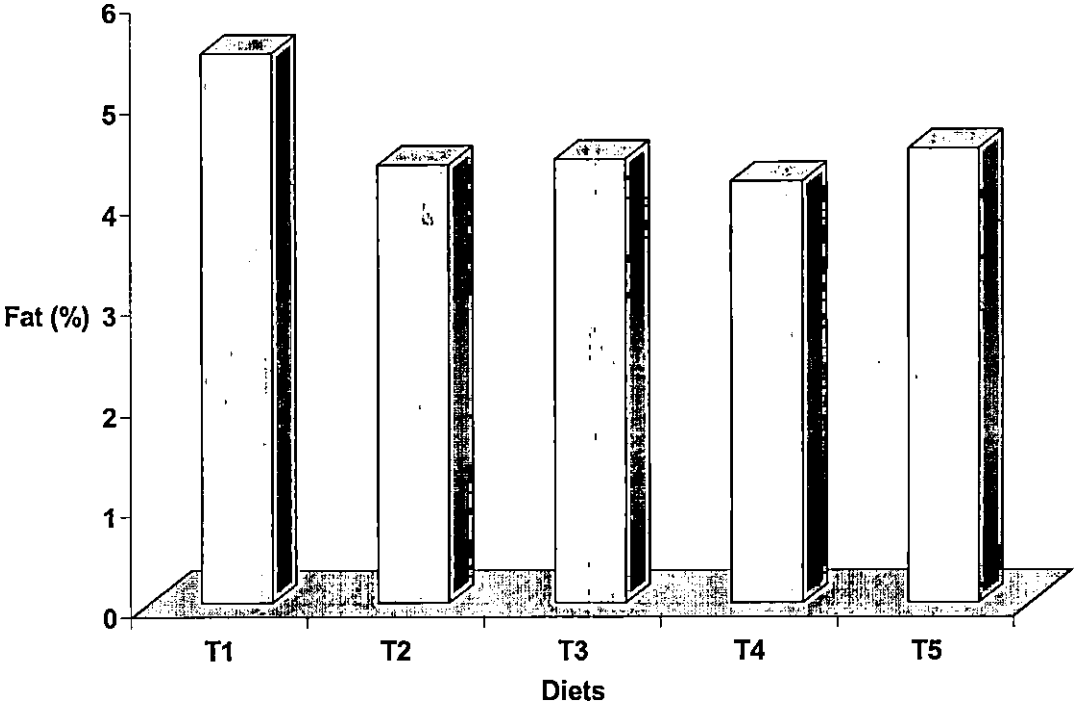

Fig.10.Final fat content in the body of *M.rosenbergii* at the end of the experiment

DISCUSSION

5. DISCUSSION

5.1. PROXIMATE ANALYSIS OF FORMULATED

Proximate composition of experimental diets was analyzed to evaluate the nutrient status. Most of the previous workers have reported that 30-35% protein may be optimum for the growth and survival of *Macrobrachium rosenbergii* post larvae. According to Balazs and Ross (1976) a protein level above 35% may be required for maximum growth in *Macrobrachium rosenbergii* juveniles. D'Abramo and New (2000) estimated a dietary protein requirement of 30-35 % crude protein for *Macrobrachium rosenbergii* post larvae. But Clifford and Brick (1979) found that a protein level of 35% was optimum for best growth and survival of *Macrobrachium rosenbergii*. Sick and Millikin (1983) estimated the protein requirement of early juvenile *Macrobrachium rosenbergii* to be around 40% and of larger prawns to be 25-30%. Protein levels in commercial feeds were 23.8 to 38.5 % in Hawaii (Corbin et al., 1983), 28 to 36 % in Taiwan (Hsieh et al., 1989) and 25 to 37 % in Thailand (Department of Fisheries, 2004). Proximate analysis of the diets in the present study revealed that they contained crude protein in the range of 33.74 to 34.99 % which falls within the levels of protein in the feeds suggested by various investigators (El-Sayed, 1997; Boonyaratpalin and Chittiwan, 2003).

Carbohydrate was in the range of 41.751 to 44.913 % in the experimental diets. Shrimps and prawns appear to utilize complex carbohydrates more efficiently than simple ones (New, 1976). Fair et al. (1980) reported that dietary fibre levels up to 30% do not appear to suppress growth in *Macrobrachium rosenbergii*. Briggs (1991) successfully employed a series of carbohydrate sources at 40% inclusion level in semi purified diets for post larvae of *Macrobrachium rosenbergii*. The prawns are known to utilize as high as 30% dietary fibre (Mitra et al., 2005).

Hilton et al. (1984) and Reigh and Stickney (1989) reported that total lipid content in the diet should not exceed 10% for *Macrobrachium rosenbergii*. In commercial feeds of *Macrobrachium rosenbergii*, a lipid level of 6 to 9% has been reported in Thailand (ASEAN/UNDP/FAO, 1988), 5 to 8 % in French Guiana (IFREMER, 1989) and 2 to 4% in Taiwan (Hsieh et al., 1989). The lipid content of the diets used in the present study was in the range of 4.2 to 5.2% which is almost the same as employed in various commercial grow out feeds of *Macrobrachium rosenbergii*.

5.2. WATER STABILITY OF FORMULATED DIETS

There was not much variation in the water stability of the diets even after four months. Water stability was found to be good after six hours with retention over 80%. In the present study, higher water stability was obtained in all the test diets. The higher water stability of the feeds, obtained in the present study may be due to the better gelatinization of starch.

5.3. EVALUATION OF EFFICIENCY OF EXPERIMENTAL DIETS

The present study was carried out to find out the feasibility of using water spinach leaves as a low cost plant protein source and to evaluate the nutritional quality of the formulated diets through growth trials employing post larvae of *Macrobrachium rosenbergii*. Evaluation of efficiency of the diets was based on growth (net weight gain), specific growth rate, survival, FCR, FCE, PER and crude digestibility coefficient. Initial as well as final carcass composition was also conducted to evaluate the test diets.

5.3.1. Growth

Among the different formulated feeds tested in the present study, the treatment T2 with *Ipomoea* leaf meal at an inclusion level of 12% and treatment T3 with *Ipomoea* leaf meal inclusion level of 24% gave superior growth performance in comparison to other experimental diets tested individually. Of the two test diets T2 and T3, the diet T2 with *Ipomoea* leaf meal inclusion of 12 % produced highest net weight of 603.63 mg, closely followed by T3 with 24

% *Ipomoea* leaf meal (net weight gain of 568.73 mg). Growth rate obtained with test diet T4 having 36% inclusion of *Ipomoea* was lower than control diet T1. Lowest net weight gain was recorded for the diet T5 (*Ipomoea* inclusion level of 48%), though not significantly different from diet T4.

Early works on shrimp and prawn by Deshimaru and Shigueno (1972), New (1976) and Conklin et al. (1977) have shown that a mixture of two or more protein sources invariably gave better growth than single protein source. The improved performance of mixed diet was mainly because of the fact that single protein source may not be able to provide all the essential amino acids in adequate levels and the deficiency may be overcome by mixing a number of protein sources so as to formulate a diet which closely meets the amino acid requirement of the test species (Boonyaratpalin and Chittiwan, 2003).

Several reports are available on the use of leaf protein as a source of protein in feed, replacing the expensive fish feed ingredients. Santiago et al. (1988) incorporated *Leucena* leaf meal at the level of 65% in the diets of Nile tilapia showed no adverse effect on growth. Cassava leaf meal (*Manihot esculanta*) was used as a protein source in the diets of Tilapia at 60% (Wee and Ng, 1986). *Leucaena* leaf meal, when it replaced fish meal in the diets of *Cyprinus carpio* showed better FCR value than commercial feed (Swamy and Devaraj, 1995). Mondal and Ray (1996) observed that, diet contained 20% inclusion of the terrestrial macrophyte Acacia in the diet of *Labeo rohita* fingerlings gave best results of growth in terms of FCR.

Richter *et al.* (2003) suggested that Moringa leaf meal can be used to substitute up to 10% of dietary protein in Nile tilapia without reduction in growth. Similarly Aneykutty *et al.*(1994) demonstrated that dried *Azolla* powder may prove to be partial protein source in the feeds of *Etroplus suratensis* and its inclusion in the diet can replace about 25% of fish meal based diet. There are a number of aquatic weeds have been used in fish feeds replacing the conventional ingredients (Patnaik *et al.,* 1991; Das *et al.,* 1994; Mohanty and Dash, 1995; Ray and Das, 1995). Their results showed that the diet with 60% inclusion level of aquatic weed

meal (*Otellia, Nymphoides, Pisia, Lemna and Azolla*) gave the highest protein and lipid deposition. *Azolla* based diets when fed to tilapia and grass carp showed a good increase in fish yield (Mohanty and Dash, 1995). Guru and Patra (2007) showed that the use of aquatic weed *Eichhornea crassipes, Lemna minor and Pistia stratiotes* in the diet of *Labeo rohita* yields positive results in the growth performance and therefore would no doubt reduce the cost of input while serving as a way of making the most discarded, the most useful.

Haniffa *et al.* (2002) studied the effectiveness of dietary plant protein sources such as *Hydrilla verticellata, Chara fragilis* and drumstick leaves on the growth of freshwater prawn, *Macrobrachium idea*, showed poor growth. Studies on *Macrobrachium rosenbergii* post larvae fed on *Spirulina* based diet also showed that the percentage weight gain was significantly lower in *Spirulina* based diet than casein diet (James et al., 1990).

In the present study, a similar observation was obtained in diets with high inclusion of water spinach (36 % and 48%). On the other hand diets with lower water spinach inclusion (12% and 24%) gave good results.

Saponins, which are found in many of the potential alternative plant derived feed sources, are considered to have a detrimental effect on fish. The negative effect of saponins might be because of their well-known effect as a surface-active component on the biological membrane by which the permeability of the intestinal mucosal cells is increased and the active nutrient transport hindered (Johnson *et al.*,1986), even though at low dietary levels, they have been shown to increase growth in tilapia (Francis *et al.*,2001). This is in agreement with the present study of utilization of water spinach leaf meal as an alternative protein source in the feed of *Macrobrachium rosenbergii* post larvae.

5.3.2. Specific growth rate

Specific growth rate can be considered as an index of growth in the evaluation of diets since it is a more refined and improved growth index than absolute weight gain or percentage growth rate (Hepher, 1988). The results of the present study indicated the highest SGR with

the diet T2 containing *Ipomoea* inclusion level of 12 %, closely followed by T3 with *Ipomoea* inclusion of 24%. The diet T1 (control) produced significantly lower SGR than the former two diets. The diets T4 and T5 with *Ipomoea* inclusion of 36% and 48 % produced lower SGR values than the diets T1.

Haniffa *et al.* (2002) obtained SGR of 0.239 for *Macrobrachium idea* juveniles fed with *Hydrilla verticellata* based pelleted diets. The authors reported that the poor performance may be due to the presence of high fibre content, imbalance of essential amino acids and low digestibility. Hari and Kurup (2003) found that there was no significant difference in growth performance in juveniles of *Macrobrachium rosenbergii* fed with equal proportions of plant and animal protein. But, the SGR values (3.8) reported in case of *Labeo rohita* fed with *Nymphoides cristatum* based pelleted diets (Patra *et al.,* 2002) is in agreement with the SGR values obtained in the present study.

The SGR obtained in the present study indicated that diets with lower inclusion levels of *Ipomoea* (12% and 24%) enhanced specific growth rate (3.6 and 3.7) whereas diets with higher inclusion levels of *Ipomoea* significantly depressed growth performance (2.7) in *Macrobrachium rosenbergii* post larvae. The results showed that the inclusion of plant protein, derived from *Ipomoea* leaves, up to 24% inclusion level had beneficial effects on the growth performance of *Macrobrachium rosenbergii* post larvae as compared to the control diet (T1). The depression of growth performance and growth parameters could likely be attributed to several factors, among which the presence of antinutrients could not be ruled out. A further possible reason for low growth at high inclusion level of *Ipomoea* might be the levels of cell wall constituents (Neutral detergent fibre and Acid detergent fibre) as reported by (Richter *et al.*, 2003).

5.3.3. Survival

The postlarval prawns of all the five treatments in the present study showed fairly good survival rates ranging from 77.5% to 82.5%, suggesting that different level of inclusion of

Ipomoea meal by replacing clam meat did not produce much variation on survival rates. New (1976), in his review on the nutritional studies of shrimps and prawns has opined that mortalities in nutritional studies are rare, unless the diet is grossly deficient in nutrients.

Neeraja (1998) found that the juvenile prawn showed fairly good survival rates ranging from 86.67% to 96.67%, suggesting that the different protein sources tested either individually or in various combinations did not produce much variation on survival rates.

Behanan *et al.* (1992) reported a survival of 76% in *M.rosenbergii* post larvae with the diet based on cat fish meat, prawn head, gluten and clam meat based diet. Similarly Raje and Joshi (1992) obtained survival rates of 66% (clam meat + fish meat based diet) to 69% (crab meat +fish liver based diet) in *M.rosenbergii* larvae. Haniffa *et al.* (2002) reported a survival rate of 86% in *M. idae* fed with *Hydrilla* based diet.

In the present study, the survival rate was between 77.5% and 82.5% which was closely similar to the result obtained by Haniffa *et al.* (2002). There was no evident feed related mortality, the reduction in survival was due to handling stress during sampling, suggesting that the different level of inclusion of *Ipomoea* leaf meal did not produce much variation on survival rates.

5.3.4. Food conversion ratio and food conversion efficiency

Food conversion ratio is the ability with which an animal can convert food for the growth process and is reflected in the ratios of food consumed to the live weight it has gained. Thus higher efficiency in food utilization indicates lower food conversion ratios. In the present study, lower food conversion ratios were registered for T2 and T3 diets, with *Ipomoea* inclusion of 12% and 24% respectively. Control diet (T1) with clam meat gave FCR higher than the former two. Diets T4 and T5 was not seen to be significantly different.

According to Colvin (1976), protein source that is deficient in essential nutrients inadequate quantities produces less efficient feed conversion ratio. In the present study, the

better food conversion ratios obtained for diets containing 12% and 24% level of inclusion of *Ipomoea* might be due to the fact that even at lower diet intake level, combined animal and aquatic weed leaf meal sources provided essential nutrients in required proportion for better growth of prawn compared to the clam meat based control diet.

Indrajasmine (1996) used plant protein (40 to 48%) and obtained FCR ranging between 1.46 and 2.25 for *Penaeus indicus*. Hari and Kurup (2003) recorded FCR values of 2.6 in *M.rosenbergii* post larvae fed with diets having a plant to animal ratio of 1:1. James et al. (1990) reported a higher FCR of 6.82 in *M.rosenbergii* post larvae fed with solar dried *Spirulina* as a protein source at 40% protein level. Similarly Haniffa *et al.* (2002) found a higher FCR of 5.11 in *M.idae* fed with *Hydrilla verticellata* based diets. However, the lower FCR values (1.1 to 2.2) obtained with present study indicated the higher efficiency in food utilization. Harpaz and Schmalbach (1986) also attributed the better efficiency of the diet supplemented with plant protein sources like leaves, to the better availability of nutrients like vitamin C to the prawns.

5.3.5. Protein efficiency ratio

Protein efficiency ratio (PER) is used to evaluate the quality of dietary protein, those with high protein efficiency ratio can be considered as better quality and those with low values as poor quality. In the present study it was found that ,*Macrobrachium rosenbergii* post larvae fed on diets containing 12% *Ipomoea* was more efficient in converting dietary protein with PER of 2.998,followed by those fed on diets with24% *Ipomoea* with PER of 2.782 . There was significant difference ($p<0.05$) in PER between these two diets. Control diet (T1) gave lower PER value compared to the former two diets. The diet T4 was not significantly different from T5.

Richter *et al.* (2003) showed better PER of 2.3 to 2.8 when Nile tilapia was fed with *Moringa* leaf meal based diets. James et al.(1990) found a PER value of 0.36 in *M.rosenbergii* post larvae fed with *Spirulina fusiformis* based diet and Haniffa *et al.*(2002)

reported a PER of 0.32 for *M.idae* juveniles fed with *Hydrilla* based diets. On the contrary, results of the present study showed higher PER values (1.5 to 3) in *Ipomoea* based diets, of which diets T2 and T3 (12% and 24% *Ipomoea*) exhibited the highest PER value (2.99 and 2.78).

DeSilva and Davy (1990) opined that for optimum growth of prawn, it is important to have a balance between dietary protein and energy ratio in diets and the ratio may differ according to the type of ingredients. The protein energy ratio lower or higher than optimum may lead to the reduction in growth of prawn (Akiyama, 1992).

In view of the above ,it can be demonstrated that the diets (T2 and T3) containing low *Ipomoea* inclusion levels of 12% and 24% might have the required protein energy levels in the diets, yielded best results and this formulation could be used to reduce the feed cost and enhanced protein conversion for *M.rosenbergii*.

5.3.6. Protein digestibility coefficient

The digestibility obtained for the test diet T2 and T3 was considerably high, when compared to the other test diets (T1, T4 and T5), indicating that incorporation of *Ipomoea aquatica* leaf meal beyond a level of 36% impairs digestibility. Better food conversion ratio and protein efficiency ratio of a diet result from better protein digestibility and absorption. The higher values obtained for the above two parameters for diet T2 and T3 directly imply that the protein digestibility and absorption were significantly better for this diet.

Maynard and Loosli (1978) have pointed out that difference in digestibility arises from incomplete digestive action or lack of complete absorption.

James *et al.* (1990) found a protein digestibility coefficient of 97.28 in *M.rosenbergii* post larvae fed with *Spirulina fusiformis* as a protein source and this high protein digestibility coefficient was due to the better utilization of *Spirulina* by *M.rosenbergii* post larvae because there was no cellulose cell wall, since *Spirulina* is a blue green alga.

But in the present study, the protein digestibility coefficient obtained in *M.rosenbergii* post larvae fed with *Ipomoea* based diet was lower than the above.

5.4. WATER QUALITY PARAMETERS

5.4.1. Temperature

Water temperature is an important factor which influences the survival and growth of any organism. *Macrobrachium rosenbergii* can survive in a wide range of temperature (18-33°c) with out any deleterious effect, provided temperature fluctuations are not severe, sudden and of long duration (Farmanfarmaian and Moore,1980). New (1990) also reported that *Macrobrachium rosenbergii* adults can tolerate wide range of 18 to 34°c, while for larvae the optimum range is 26 to 31°c. Temperature <14°c or >35°c are reported to be lethal for post larvae, optimum being 29 to 31°c. The weekly range of temperature observed during the present experimental period was 26.1 to 30.3°c. The values recorded in the present study were with in the optimum range suggested for the growth of *Macrobrachium rosenbergii* post larvae.

5.4.2. pH

pH of water is another factor which has been reported to affect the growth of prawns. In the present study, pH of water was almost uniform in all experimental tanks and varied between 6.9 to 8.4. New and Singholka (1982) and Sandier and Smith (1985) reported a pH range of 7.5 to 8.5 as optimum for culture of *Macrobrachium spp.* Malecha *et al.* (1980) and Sandifer and Smith1985) observed that high pH values were not favorable for the growth of *Macrobrachium rosenbergii.* Ideal pH for *Macrobrachium rosenbergii* is reported to be 7 to 8.5 by Hsieh *et al.* (1989). pH of water in the present study was within the ideal range.

5.4.3. Dissolved oxygen

Temperature of water and metabolic rate of prawn influence the physiological need for oxygen. New and Singholka (1982) reported that an oxygen concentration of 75% saturation was optimum for the growth of *Macrobrachium spp.* Vasaquez *et al.* (1989) found that the optimum level of dissolved oxygen in pond conditions for *Macrobrachium* culture is 6 to 8 ppm. During the present study, weekly dissolved oxygen values in the experimental tanks ranged from 6.3 to 8.39 ppm, since mild aeration was provided to the tanks. These values were found to be optimum for rearing of *Macrobrachium rosenbergii* post larvae.

5.3.5. Total alkalinity

Natural waters that contain 40mg/l or more of total alkalinity are considered more productive than water of low alkalinity. Waters of low alkalinity are poorly buffered against fluctuations in pH and consequently rapid reduction in pH occurs when carbon dioxide levels goes down. Ayyappan and Rao (2000) reported tolerable range of alkalinity as 40 to 150 ppm. Total alkalinity values recorded in the present study ranged from 90 to 120 ppm which was in this tolerable limit.

5.4.5. Ammonia

In the present study the ammonia concentration ranged between 0.056 mg/l to 0.089 mg/l.

Growth of *Macrobrachium rosenbergii* is found impaired at unionized ammonia concentration of 0.1mg/l or higher (Ayyappan and Rao, 2000). The concentrations of ammonia recorded in the present study were within these limits.

5.5. CARCASS COMPOSITION OF POST LARVAE OF *M.ROSENBERGII*

Carcass analysis of experimental animals was done initially and on termination of the study. From the result it was clear that prawns fed with a diet containing optimum level of inclusion of *Ipomoea aquatica* leaf meal showed proper utilization of feed and protein. The level of inclusion of *Ipomoea aquatica* leaf meal was significantly influenced the chemical composition of body of prawns.

5.5.1. Protein

The results showed that protein content of the test animals increased with increase of *Ipomoea* leaf meal in the diet and protein reached maximum at 24% inclusion of *Ipomoea* followed by diets T5 andT4. Richter et al. (2003) also reported similar increase of protein in the body of Nile tilapia fed with *Moringa* based diets. It is thus clear from the present study that better protein content of the *Ipomoea* leaf meal is responsible for the higher content of protein in the body of *Macrobrachium rosenbergii* post larvae.

5.5.2. Lipid

Lipid content in the whole body of the prawns at the end of the feeding experiment were estimated and found that the treatments except control (T1) and T4 were not significantly different. The lipid content of prawn fed with control diet is superior to other diets. This shows that better utilization of lipid was from clam meat based diet compared to aquatic weed based diet.

5.5.3. Moisture

The level of whole body moisture was significantly lower in *Ipomoea* based diets compared to the control diet (T1). The moisture content of the body is found to be decreased with increased level of inclusion of *Ipomoea* leaf meal.

5.5.4. Carbohydrate

From the present experiment it was found that carbohydrate content of body of experimental animal was maximum for T5 with 48 % Ipomoea inclusion and minimum for T1 without *Ipomoea* inclusion.

SUMMARY

6. SUMMARY

An experiment has been carried out to evaluate the efficacy of pelleted feeds formulated by replacing clam meat with *Ipomoea* leaf meal. Clam meat was progressively replaced with *Ipomoea* leaf meal to *Macrobrachium rosenbergii* post larvae at various levels. A comparison has also been made between control diet and diets containing *Ipomoea* leaf meal in various inclusion levels.

In the present study post larvae of *M.rosenbergii* having an average weight of 0.077g were used as experimental animals. The experiment was conducted in Completely Randomized Design with five treatments and 4 replications each, for a period of 60 days.

Proximate analysis of various feed ingredients used in the formulation of test diets showed that dried clam meat contained highest crude protein of 50.7% followed by ground nut oil cake (42%) and *Ipomoea* leaf meal (24.9%).

Five isonitrogenous test diets were prepared for the study. They were diet T1 with clam meat+GOC+tapioca+Wheatbran+sunflower oil+ vitamin mineral mix, diets T2,T3,T4 and T5 with clam meat replaced with 12%, 24%, 36% and 48% *Ipomoea* leaf meal respectively.

Proximate analysis of the formulated diets showed that crude protein content of the diets ranged between 33.37% and 34.99%, crude fat content ranged between 4.2% and 5.2%.

The various observations in the water quality parameters were found to be well within the tolerable limits for the optimum growth of *M.rosenbergii* post larvae.

Various evaluation indices viz., net weight gain, specific growth rate, percentage survival, FCR, FCE, PER, protein digestibility coefficient and carcass composition of the test animals were determined.

On termination of 60 days rearing, better growth rates were recorded in *M.rosenbergii* post larvae fed with 12% inclusion of *Ipomoea* leaf meal.

Higher specific growth rates were recorded in prawn post larvae fed on diets T2 and T3 containing 12% and 24% inclusion of *Ipomoea* leaf meal than control diet without *Ipomoea.*

The survival rate of *M.rosenbergii* post larvae was not found to be influenced substantially by various inclusion levels of *Ipomoea* leaf meal and the survival rate recorded in various treatments ranged between 82.5% and 77.5%.

Low food conversion values were obtained in prawns fed on the diets with 12% and 24 % level of inclusion of *Ipomoea* leaf meal over the control diet without *Ipomoea* leaf meal.

High protein efficiency ratios were recorded with diets T2 (12% *Ipomoea*) and T3 (24% *Ipomoea*) based on *Ipomoea* leaf meal.

REFERENCES

7. REFERENCES

Agbede, S.A.and Falaye, A.E.1997. Nutritive value of three aquatic weeds as diets for Nile tilapia (*Oreochromis mossambicus*) *J.Trop. Resources*. 4(1): 19-23

Akiyama, D. M., Dominy, W.G. and Lawrence, A.L. 1992. Penaeid shrimp nutrition. *Marine shrimp culture: Principles and practices* (eds.Fast, A.W. and Lester, L.J.L). *Elsevier science Publishers*. Amsterdam. pp.535-568

Alava, V.R. and Pascual, F.P. 1987.Carbohydrate requirements of *Penaeus monodon* (Fabricius) juveniles. *Aquaculture*. 61: 211-217

Alceste, C.2000. Tilapia: alternative protein sources in tilapia feed formulation. *Aquaculture*. 26: 14-22

Ali, S.A. 1998. Studies on the evaluation of different sources of protein, carbohydrate and mineral requirements for juvenile prawn *Penaeus indicus* H.Milne Edwards. Ph.D. Thesis, Cochin University of Science and Tecnology,Cochin, 243 p.

Aneykutty, Sherief, P.M. and James, T.1994. Effect of different dietary inclusion levels of *Azolla pinnata* on the growth, food conversion and muscle composition of *Etroplus suratensis* (Bloch). *J.Aquaculture trop*. 9: 87-94

Ang, K.J., Lee, Y.M. and Law, A.T.1992. Protein requirement for broodstock *M.rosenbergii*. *Third Asian Fisheries Forum*.26-30 October 1992.Singapore.*Asian Fisheries Soc*.Manila; Philippines.Abstr: 8

Anilkumar, P. 1994. Evaluation of different protein sources in the formulated feeds for *Macrobrachium rosenbergii* (de Man). M.F.Sc. Thesis submitted to Kerala Agricultural University, College of Fisheries, Panangad, Cochin, India. P.121

*Antiporda, J.L. 1986. Optimum protein requirements for *Macrobrachium rosenbergii* juveniles. Working paper, NACA/WP/86/45. *Network of Aquaculture centres in Asia,* Bangkok, Thailand. 20 p.

AOAC.1990. Methods of Analysis. *Association of Official Analytical Chemists.* 15th ed., Arlington, Virginia, 1298 p.

AQUACOP.1990.Freshwater prawns. *Aquaculture, I* (ed.Barnabe, G.). Ellis Haqwood Ltd, England. pp. 501-528

AQUACOP. 1976. Incorporationde proteins vegetables dans unaliment compose pour *Macrobrachium rosenbergii. Aquaculture.8:71*-80

*ASEAN/UNDP/FAO.1988.Country review paper (Thailand) presented at ASEAN Workshop on shrimp and finfish feed Development, 25-29 October. JohoneBabru, Malaysia. *ASEAN/UNDP/FAO* Regional small scale coastal fisheries Development project, Manila, Philippines. pp.13-15

Ashmore, S.B., Stanley, R.W., Moore, L.B. and Malecha, S.R. 1986. Effect of growth and apparent digestibility of diets varying in grain source and protein level in *Macrobrachium rosenbergii. J.Wld.MaricultureSoc.* 16: 205-216

Ayyappan, S. and Piliai, N.G.K. 2005. Indian Fisheries in Global Context, *Indian Farming.*55 (7):16-24

Ayyappan, S. and Rao, J. 2000. Hatchery technology of Giant freshwater prawn, *Macrobrachium rosenbergii. Fishing chimes.*19 (10 &11): 112-121

Bairagi, A., Ghosh, K.S., Sen, S.K., Raj, A.K. and Sarkar Ghosh, K.2002. Duck weed leaf meal as a source of feedstuff in formulated diets for rohu fingerlings after fermentation with a fish intestinal bacterium. *Bioresource Technology* 5(1): 17-24

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

Balazs, G.H. and Ross, E. 1976. Effect of protein source and level on growth and performance of the captive freshwater prawn, *Macrobrachium rosenbergii. Aquaculture.* 7: 299-313

Basudha, C.H. and Vishwanath, W.1997. Formulated feed based on aquatic weed *Azolla* as fish meal for rearing medium carp *Osteobrahma belangeri* (*Valenciennes*). *J.Aquaculture in Trop.* 12: 155-164

Behanan, L., Mukundan, M.R., Alphi, K. and Sherief, P.M. 1992. Formulation and evaluation of particulated fed for *Macrobrachium rosenbergii* post larvae (ed. Silas, E.G.). *Fresh water prawns. Proc.Natn.Symp.Fresh water prawn (Macrobrachium spp.) held at Faculty of Fisheries*, K.A.U. 1990cochi, India, Dec 12-14, 1990.pp.238-240

Behanan, L. and Mathew, S.1995. Significance of formulated feeds in post larval rearing of fresh water prawn. *Fishing chimes.* 15(3):39-41

Bindu, R.P., Tripathy, A.P.and Dandapat, J. 2001. Comparative evaluation of biochemical parameters in wild and cultured broodstock of *Macrobrachium rosenbergii. Indian J.Fisheries.*48 (1): 35-39

Boghen, A.D., Castell, J.D.and Conklin, D.E. 1982. In search of a reference protein to replace 'Vitamin free' casein in lobster nutrition studies. *Can.J.Zool.* 60: 2033-2038

*Boonyaratpalin, M.and New, M.B. 1980. Evaluation of diets for giant freshwater prawn reared in concrete ponds. *Thai fish. Gaz.* 33 (5): 555-561

Boonyyaratpalin, M.and Chittiwan, V.2003. *Macrobrachium* Nutrition: feed and feeding. Fresh water prawns: Advances in biology, aquaculture and marketing (eds. Nair,C.M,Namboodiri,D.D.,Jose,S.,Sankaran,T.M.,Jayachandran,K. and Salin,K.R). pp. 521-534

Bordner, C.E.1989. A standard reference diet for crustacean nutrition research. Growth and survival of juvenile Dungeness crabs, *Cancer megister. J.Wld .Aquaculture.Soc.* 20(30) : 119-121

Briggs, M.R. 1991. The performance of juvenile prawns, *Macrobrachium rosenbergii*, fed a range of carbohydrate sources in semi purified diets. *J.Wld. Aquaculture.Soc.* 22 (3): 16

Briggs, M.R.P., Jauncey, K. and Brown, J.H. 1988.The cholesterol and lecithin requirements of juvenile prawn *Macrobrachium rosenbergii* fed semi purified diets. *Aquaculture.* 70: 121-129

Castell, J.D.1983. Fatty acid metabolism in crustaceans. Proceedings of the 2nd International Conference on aquaculture nutrition. World Aquaculture Society. Baton Rouge, Louisana, USA. Pp.124-145

Cavalli, R.O., Lavens, P. and Sorgeloos, P.1999. Performance of *M.rosenbergii* broodstock fed diets with different fatty acid composition.*Aquaculture.*179: 587-402

Cavalli, R.O., Menschaert, G., Lavens, P. and Sorgeloos, P.2000. Maturation performance, Offspring quality and lipid composition of *M.rosenbergii* females fed increasing levels of dietary phospholipids. *Aquaculture Int.*8 (1): 41-58

Cavalli, R.O., Tamtin, M., Lavens, P. and Sorgeloos, P.2001. Variation in lipid classes and fatty acid content in tissues of wild *Macrobrachium rosenbergii* (de Man) females during maturation. *Aquaculture*.193: 311-324

Chamberlain, G.W.1988. Stepwise investigation of environmental and nutritional requirements for reproduction of penaeid shrimp. Ph.D. Thesis, Department Wildlife and Fisheries Science, Texas A &M University, Texas, USA

Clarke, A., Brown, J.H. and Holmes, L.J.1990. The biochemical composition of eggs from *Macrobrachium rosenbergii* in relation to embryonic development. *Comparative Biochemistry and Physiology*. 96: 505-511

Clifford, H.C. and Brick, R.W. 1979. A physiological approach to the study of growth and bioenergetics in the fresh water shrimp *Macrobrachium rosenbergii. J.Wld. aquaculture Soc.* 10: 701-719

Colvin, P.M. 1976. Nutritional studies on penaeid prawn: Protein requirements in compounded diets for juvenile *Penaeus indicus. Aquaculture.* 7: 315-326

Conklin, D.E., Devers, K. and Bordner, C.E. 1977. Development of artificial diets for lobster *H.americanus. Proc.Ann.Meet Wld. Mariculture Soc.*8: 841-855

Corbin, J.S., Fujimoto, M.M. and Iwaai, T.Y.1983. Feeding practices and nutritional considerations for *Macrobrachium rosenbergii* culture in Hawaii (ed. McVey, J.P.). *CRC Handbook for mariculture*, volume I. Crustacean aquaculture CRC Press, Inc.Boca Raton.pp. 391-412

Cowey, C.B. and Forster, J.R.M. 1971. The essential amino acid requirements of the prawn *Palaemon serratus*. The growth of prawns on diets containing proteins of different amino acid compositions. *Mar.Biol.*10: 77-81

Cruz-Rique, E.L., Guillaume, J., Cuzon, G. and AQUACOP.1987. Squid protein effect on growth of four penaeid shrimp.*J.Wld.Aquaculture.Soc.* 18: 209-217

Cruz-Suarez, L.E., Marie, D.R., Vega, J.A.M. and Ebeling, P.W. 1993. Evaluation of two shrimp by-products meal as protein source in the diet of *Penaeus vannamei. Aquaculture.*115: 53-62

Cruz-Suarez, L.E., Antimo-Perez, J.S., Luna-Mendoza, N. and Ricque-Marie, D.2001. Effect of two different vegetable/animal protein proportions on the optimum dietary protein /energy ratio for *Littopenaeus vannamei* and *L.stylirostris* growth. *Wld. Aquaculture .Soc.*USA.21-25 Jan2001.148 p.

D'Abramo, L.R. and Reed, L.1988. Optimal dietary protein level for juvenile fresh water prawn *Macrobrachium rosenbergii. J.Wld. Aquaculture Soc.*19 (1): 25

D'Abramo, L.R. and New, M.B.2000. Nutrition, feeds and feeding. Freshwater prawn culture (eds. New, M.B. and Valenti, W.C.).The farming of *Macrobrachium rosenbergii. Blackwell Science.*pp. 203-220

D'Abramo, L.R. and Sheen, S.1993. Polyunsaturated fatty acid nutrition in juvenile fresh water prawn *Macrobrachium rosenbergii.Aquaculture.*115: 63-86

D'Abramo, L.R., Moncreiff, C.A., Holcomb, F.P., Montanez, J.I, and Buddington, P.R. 1994. Vitamin C requirement of the juvenile fresh water prawn, *Macrobrachium rosenbergii. Aquaculture.128: 269-*275

D'Abramo, L.R., Daniels, W.H., Fondren, M.W .and Brunson, M.W.1995. Management practices for the culture of the freshwater prawn (*M.rosenbergii*) in temperate climates. Mississippi Agricultural and Forestry Experimental station *Tech.Bulletin* No: 1030

Dalfiah, Kabangnga, N. and Palinggi, N.N.1998. Shrimp head meal as an animal protein source for feed Mangrove crab *Scylla serrata. J.Indonasia.* 43 (3) p. 45-49

Dall, W. and Smith, D.M.1987. Changes in protein bound and free amino acids in the mussel of the tiger prawn *Penaeus esculentus* during starvation. *Marine Boil.* 95: 509-520

Daniels, W.H., Cavalli, R.O.and Smullen, R.P.2000. Broodstock Management. Freshwater prawn culture (eds.New, M.B. and Valenti, W.C.). The farming of *Macrobrachium rosenbergii. Blackwell Science.* pp. 41-51

Das, D., Sikdar, K. and Chotterjee, A.k.1994. Potential of *Azolla pinnata* as biogas generator and as a fish feed. *Int. J. Environment and health.*36 (3): 186-191

Das, K.M., Mukhopadhya, P.K. and Nanda, S.k.1995. Evaluation of four formulated diets in *Macrobrachium rosenbergii* juveniles. *J. Aquaculture Trop.*10 (2): 129-137

Das, N.N., Saad, C.R., Ang, K.J., Law, A.T. and Harmin, S.A.1996. Diet formulation for *Macrobrachium rosenbergii* (de Man) broodstock based upon essential amino acid profile of its eggs. *Aquaculture research.*27: 543-555

Das, N.N., Saad, C.R., Ang, K.J., Law, A.T. and Harmin, S.A.1993. Some aspects of the protein requirement for broodstock *Macrobrachium rosenbergii* (de Man). *Int.Conf.Fisheries and the Environment: Beyond2000.Serdang, selangor, Malaysia.6-9* December 1993

De Silva, S.S. and Davy, F.B.1990.Fish nutrition research for semi-intensive culture system in tropics –some strategies for the future. *J.Wld.Aquaculture Soc.*17: 37-43

Del-Carmen-Gonzalez-Pena, M., Gomes, S.Z.and Moriera, G.S. 2001. Effects of Dietary Fibers on Growth and Gastric Emptying Time of the Fresh water Prawn *Macrobrachium rosenbergii* (de man, 1879). *J. Wld. Aquaculture Soc.* 33: 441-447

Department of Fisheries, 2004. *Quality Guideline for Shrimp Feed Registration in Thailand.* Feed quality control section Thailand, 5 p.

Deshimaru, O. and Shigueno, K.1972. Introduction to artificial diet for prawn, *Penaeus japonicus. Aquaculture.*1: 115-133

Deshimaru, O.1982. Protein and amino acid nutrition of the prawn *Penaeus japonicus*. (eds. Pruder, G.P., Langdon, C.J. and Conklin, D.E.). *Proc.second Int.Conf. Aquaculture Nutr.Biochemical and physiological approaches to shell fish nutrition.* Louisiana State University, Baton Rouge, Lousiana.pp.106-123

Devaraj, K.V. and Keshavappa, G.Y.1981. The use of poultry dropping as an ingredient in fish feed. *Proc.of seminar on some aspects of inland aquaculture in Karnataka.*14-15 July 1980.College of Fisheries, Manglore.pp. 87-93

*Dokken, Q.R. and Lawrence, A.R. 1985. Growth response of *Penaeus vannamei* to feeding frequency and dietary ingredients, soyabean meal, white fish meal mixture, squid meal and squid puree. *Wld. Mariculture Soc.* Orlando, Florida, U.S.A.

*El-Sayed, A.F.M.1997. Growth rates and feed efficiency of the freshwater prawn *Macrobrachium rosenbergii* fed varying protein and energy levels. *Bull.Nat.Inst. Ocean and Fish. Egypt.*23: 439-448

El-Sayed, A.F.M.1990. Long term evaluation of cotton seed meal as a protein source for Nile tilapia *Oreochromis niloticus.Aquaculture*.84: 315-320

El-Sayed, A.F.M.1999. Alternative dietary protein sources for farmed tilapia, *Oreochromis spp. Aquaculture*.179: 149-169

Eusebio, P.S. and Coloso, R.M.1998. Evaluation of leguminous seed meals and leaf meals as plant protein sources in diets for juvenile *Penaeus indicus .Israeli J. Aquaculture.* 50(2): 47-54

Fair, P.H., Fortner, A.A., Millikin, M.R. and Sick, L.V. 1980. The effect of dietary fibre on the growth, assimilation and cellulase activity of *Macrobrachuium rosenbergii. Proc Wld. Maricultu.Soc*.11: 369-381

Falaye, A.E., Olaniran, T.S. and Aroso, B.O. 2001. The growth and survival rate of *Oreochromis niloticus* fingerlings fed with varying percentages of *Leucaena leucocephala* leaf meal based diets. *Proceedings of the 14th Annual Conference of Fisheries Society of Nigeria, Ibadan.19*-23 January1998

Farmanfarmaian, A. and Moore, R.1980. Diseasonal thermal Aquaculture. Effect of temperature and dissolved oxygen on survival and growth of *Macrobrachium rosenbergii. Proc.Wld. Maricult.Soc.* 9: 55-56

Fasakin, E.A., Balogun, A.M. and Fagbero, O.A. 2001. Evaluation of sun-dried water fern *Azolla africana* and duck weed, *Spirodela Polyrrhiza*, in practical diets for Nile tilapia, *Oreochromis niloticus*, fingerlings. *J.Applied Aquaculture*.11 (4): 83-92

Fasakin, E.A., Balogun, A.M. and Fasuru, B.E.1999.Use of duck weed, *Spirodela polyrrhiza L. Schleiden*, as a protein feedstuff in practical diets for tilapia, *Oreochromis niloticus L.Aquacult.Res*.30: 313-318

Felix, N. and Prince Jayaseelan, M.J.2006. Effects of different protein diets on growth and food conversion ratio of post larvae of *Macrobrachium rosenbergii* (de Man). *Indian J.Fish.*53 (2): 175-180

Felker, P. and Bandurshi, R.S.1977. Protein and amino acid composition of legume seeds. *J.Sci.food.Agric.*28: 791-797

Fetuga, B.L., Babatunde, G.M. and Oyenuga, V.A.1973. Protein quality of some Nigerian feed stuffs-I, Chemical assay of nutrients and amino acid composition. *J.Sci.Food Agric.*24: 1505-1514

*Forster, J.R.M. and Beard, T.W.1973. Growth experiments with the prawn *Palaemon serratus* fed with fresh and compounded foods. *Fish Invest. Ser.II.*27 (7): 1-16

Forster, J.R.M.1976. Studies on the development of the compounded diets for prawns (eds.Price, K.S., Shaw, W.N. and Danberg, K.S.).*Proc.First Int.Conf.Aquaculture Nutr.* NOAA (sea Grant). pp. 229-248

Fox, C., Brown, J.H. and Briggs, M. 1994. The nutrition of prawns and shrimp in aquaculture: a review of recent research. *Recent Advances in aquaculture* (eds. Muir, J.F. and Roberts, R.J). *Blackwell Science Ltd.*, United Kingdom. pp. 131-206

Francis, G., Makkar, H.P.S. and Becker. 2001. Effects of Quillaja saponins on growth, metabolism, egg production and muscle cholesterol in individually reared Nile tilapia (*Oreochromis niloticus).comparative Bio.Chem.Physiol.* 129: 105-114

Freuchtenicht, G.W., Bark, L.E., Malecha, S.R. and Stanley, R.W.1988. The effect of protein level in *Macrobrachium rosenbergii* individually reared in clear water flow through aquaria.

Presented at the 19th Annual Meeting of the *World Aquaculture Society*. January 2-9, 1988.Honolulu, Hawaii.

Garces, L.L. and Heinen, J.M.1989. Supplementary foods for post larval fresh water prawns, *Macrobrachium rosenbergii. J.Wld. Aquaculture Soc.* 20 (1): 39

Gomez, G.D., Nakagawa, H. and Kasahara, S.1988. Effect of dietary protein/starch ratio and energy levels on growth of the giant fresh water prawn *Macrobrachium rosenbergii. Nippon Suisan Gakkaishi.* 54: 1401-1407

Gopal, C.1986. Nutritional studies in juvenile *Penaeus indicus* with reference to protein and vitamin requirements. Ph.D. Thesis, Centre of Advanced Studies in Mariculture, CMFRI, Cochin.306 p.

Guru, S.R. and Patra, A.K.2007.Evaluation of nutritional impact of water weed-based feeds-on the growth rate of *Labeo rohita. Fishing chimes.* 27(4): 28-36

Haniffa, M.A., Sethuramalingam, T.A., Selvam, S.A. and Raj, A.J.A.2002.Effect of Plant Proteins in iso-caloric Feeds on Growth Performance of Fresh water Prawn *Macrobrachium idea* (Heller).*Fishery Technology* .39(1): 54-58

Hari, B. and Kurup, B.M.2003. Comparative dietary protein levels and plant –animal protein ratios in *Macrobrachium rosenbergii* (de Man). *Aquaculture Nutri.* 9: 131-137

Harpaz, S.and Schmalbach, E.A.1986. Improved growth and health of the Malaysian prawn, *Macrobrachium rosenbergii* by addition of fresh leaves to the artificial diets. *Aquaculture.* 55(2): 81-85

Harrison, K.E.1990. The role of nutrition in maturation, reproduction and embryonic development of decapod crustaceans-a review. *J.Shell Fish Research.*9: 1-28

Harrison, K.E.1997. Broodstock nutrition and maturation diets. Crustacean nutrition, Advances in world Aquaculture (eds. D'Abramo, L.R., Conklin, D.M .and Akiyama, D.M.). *Wld.Aquaculture Soc.* pp. 390-408

Hasting, W.E. 1976. Fish nutrition and fish feed manufacture. *FAO Tech.Conf.on Aquaculture, Japan* FIR:AQ/Conf/76/R.,23 .13 p.

Hepher, B.1988. Nutrition of pond fishes. Cambridge University Press, Cambridge. 388 p.

Hien, T.T.T. and Phu, T.Q. 2000. Effects of diet on composition and titers of yolk protein, vitellogenin in hemolymph of giant freshwater prawn. Proceeding of the National Workshop on aquaculture. Cantho University. pp. 391-394

Hien, T.T.T., Hai, T.N., Phuong, N.T., Hang, B.T.B., Yen, D.T., Ogata, H. and wilder, M.N. 2001. The effects of feeding diets containing various sources of lipid on the performance of freshwater prawn broodstock. Institute for Aquaulture and Fisheries Sciences, College of Agriculture, Cantho University, Cantho, Vietnam

Hilton, J.W., Harrison, K.E. and Stanley, J.S.1984. A semi purified test diet for *Macrobrachium rosenbergii* and lack of need for supplemental lecithin. *Aquaculture*. 37: 209-215

*Hsieh C.H., Chao N.H., Gomes L.A.O and Liao I.C. 1989. Culture practices and status of the giant fresh water prawns, Macrobrachium rosenbergii, in Taiwan. *Anais do III Simposio Brasileiro sobre Cultivo de Camarao.15-20 outubro 1989, Joao Pessoa, vol.2: Camarao de Agua Dou e Outros* (eds. Martin, M.M.R., Correia, E.S. and Cavalheiro, J.M.). MCR Aquaculture Joao Pessoa. pp.85-109

*IFREMER, 1989. Fresh water prawn *Macrobrachium rosenbergii* culture in French Overseas Territories: Origin, extention and presented at Aquaculture'89. 12-16 February, Los Angeles. pp.17

Indrajasmine, G.1992. Effect of Non-conventional Feed Ingredients on Biochemical Composition and Growth in the Indian White Prawn *Penaeus indicus*, Thesis submitted to Manonmanian Sundaranar University, Thirunelveli.

Islam,M.S.,Kabir,M.S.,Khan,S.I.,Ekramullah,M.,Nair,G.B.,Sack,R.B.and Sack,D.A.2004. Wastewater-grown duckweed may be safely used as fish feed. *Can.J.Microbiology*. 50(1): 51-56

Ittoop, G.1996. Vitamin C requirement and its deficiency syndromes in *Macrobrachium rosenbergii* juveniles. M.F.Sc.Thesis. Submitted to Kerala Agricultural University, Faculty of Fisheries, Cochi, and Kerala. pp. 30-35

Jackson, I.T., Capper, B.S. and Matty, A.J.1982. Evaluation of some plant proteins in complete diets for tilapia, *Sarotherodon mossambicus*. *Aquaculture*.27: 97-109

James T., Sherief P.M., Nair C.M and Thampy D.M. 1990. Evaluation of Spirulina fusiformis as a protein source in the diet of post larvae of Macrobrachium rosenbergii. *Fresh water prawns. Proceeding of National Symposium on Fresh water Prawns (Macrobrachium spp.)*, Kochi, 12-14 December 1990, India (ed. Silas, E.G.).Kerala Agriculral University, Thrissur, India. pp. 234-237

Jayalakshmi, B. and Natarajan, P.1993. Growth, conversion efficiency and moulting of prawn *Macrobrachium idella* fed on feeds containing plant matter. *Indian J. Mariculture Sci*.22 (2): 149-150

Jayalakshmy, B. and Natarajan, P.1994. Growth, conversion efficiency and moulting frequency of *Macrobrachium idella* fed on different artificial diets. *J. AquacultureTrop.*9 (3): 193-200

Jeyalakshmi, G., Joseph G., Irwin and Daniel, T.1997. Nutritional value of leaf protein concentrates in pelleted feed for fresh water prawn *Macrobrachium malcolmsonii.* Paper presented at National Workshop on Fish and Prawn Feeds, CIFA, Kausalyaganga, 2-3 September, 1997. Abstr: 11

Johnson, I.T., Gee, J.M., Price, K., Curl, C. and Fenwick, G.R.1986. Influence of saponins on gut permeability and active nutrient transport in vitro. *J.Nutr.* 116: 2270-2277

Josekutty, P.A. and Jose, S.1992. Growth response of *Penaeus monodon* to pelleted feeds to different protein sources. National seminar on prawn feeds, 1992. *Asian Fish.Soc.*Abstr: 21

Joseph, J.D.and Williams, J.E.1975. Shrimp head oil: A potential feed additive for mariculture. *Proc.Wld. Mariculture Soc.*6: 147-155

Kabangnga, N., Palinggi, N.N. and Sulaeman.1999. Raw material of animal protein sources in feed of mangrove crab. *Proceedings of the National Seminar on the results of agricultural technology assessment and research towards governance autonomous era, Bogor Indonasia.* pp.794-797

*Kanazawa, A.1993. Essential phospholipids of fish and crustaceans. *Fish nutrition in practice,* INRA edn.Verasailles, Cedex, France. pp. 519-530

Kanazawa, A., Paulraj, R. and Ahmed Ali, S.1982. Preparation of artificial diets for nutritional studies. *Manuel of Research methods for fish and shell fish nutrition .CMFRI.spec.publ.*8: 90-94

Kanazawa, A., Teshima, S. and Endo, M.1979. Requirements of prawn *Penaeus japonicus* for essential fatty acids. Mem.Fac.Fish., Kagoshima Univ.28: 27-33

*Kanazawa, A., Teshima, S.and Sakamoto, M.1985. Effects of dietary lipid, fatty acids and phospholipids on growth and survival of prawn (*Penaeus japonicus*) larvae. *Aquaculture*.127: 29-40

Kanazawa, A.1992. Recent advances in penaeid nutrition in Japan (eds.Allan, G.L.and Dall, W.). *Proc.Aquaculture.Nutr*. Workshop NSW Fisheries Salamander Bay, Australia. pp. 64-71

Kohler, C.C. and Kruger, S.P.1985. Use of pressed Brewer's grain as feed for fresh water prawn (*Macrobrachium rosenbergii*). *J. Wld .Maricult.Soc*.16: 181-182

Kompiang, P.1990. Shrimp nutrition. Presented at AQUATECH'90, Kaula Lumpur. p. 60-78.

Koshio, S., Teshima, S. and Kawakawa, A. 1994. The effects of protein sources and rearing methods on dietary lecithin supplement for fresh water prawns, Macrobrachium rosenbergii. *Proceeding of the 3rd Asian Fisheries Forum*, Singapore, 26-30 October 1992(eds. Chou,L.M., Munro,A.D., Lam,T.J., Chen,T.W., Cheong,L.K.K.,Ding,J.K., Hooi,K.K., Khoo,H.W., Phang,V.P.E., Shim,K.F.and Tan,C.H.). Asian Fisheries Society, Manila, the Philippines. pp. 685-688

Koshio, S., Kanazawa, S. and Teshima, S.E.1992. Nutritional evaluation of dietary soyabean protein for juvenile fresh water prawn, *Macrobrachium rosenbergii. Bull.Jap.Soc.Sci.Fish*.58 (5): 965-970

Krishnankutty, N.2005. Plant protein in fish feed. *Current Science*.88 (6): 865-867

Landesman, L., Jiayang, C., Yuri, Y. and Goodwin, J. 2002. Nutritional value of waste water –grown duckweed for fish and shrimp feed. *World Aquaculture*.33 (4): 39-40

Law, A.T., Chin, K.S.S., Ang, K.J.and Kamarudin, M.S.1990. Digestibility of low-cost ingredients in pelleted feeds by *Macrobrachium rosenbergii* (de Man) (eds.Hirana, R.and Hanyo, I.). The 2nd Asian Fisheries Forum. *Asian Fisheries Society*, Manila, Philippines. pp.333-336

Law, A.T., Poh, Y.T.and Ang, K.J.1992. Least-cost feed formulation for juvenile *Macrobrachium rosenbergii* by using linear programming technique. The 3rd Asian Fisheries Forum, 26-30 October 1992, Singapore, Asian Fisheries Society, Manila, Philippines. Abstr: 5

Lightner, D.V., Himtea, B., Magarelli, P.C.and Colvin, L.B.1979. Ascorbic acid nutritional requirement and role in wound repair in penaeid shrimp.Proc. *Wld .Maricult.Soc.*10: 513-528

Ling, S.W.1969. Methods of rearing and culturing Macrobrachium rosenbergii .Proceedings of the world conference on shrimp and prawn (Mexico), 1967: FAO Ref. Pap.E.31.FAO, Rome, pp. 607-619

Lovel, R.J.1989. Fish nutrition in Aquaculture. (ed. Adlaka, D.K.). Aquaculture Research Needs for 2000AD. Oxford IBH Publishing Co.Pvt.Ltd, New Delhi, pp: 105-122

Lovell, T.1998. Feeding penaeid shrimp Nutrition and feeding of fish.II Edition. Kluwer Academic Publishers, Boston.Dordrecht, London. pp. 227-240

Majid, F.M.1998. Aquatic weeds utility and Development.Agrobotanica, New Delhi.98 p.

Malecha, S., Sarvar, D and Orizuka, D.1980. Approaches to the study of domestication in the fresh water prawn *Macrobracghium rosenbergii* with special emphasis on the Annenue and Malaysian stocks. *Pro.Wld .Maricult.Soc.*11: 500-528

Manju, K.G. and Devendran, K.1997. Effect of bacteria and Actinomycetes as single cell protein feed on growth of juveniles of *Macrobrachium Idella* (Hilegendorf). *Indian J. Exp.Biol.*35 (1): 53-55

Marsden, G.E., McGuren, J.J. and Hansford, S.W.1997. A moist artificial diet for prawn broodstock: its effect on the variable reproductive performance of wild caught *Penaeus monodon. Aquaculture.* 149: 145-156

Maynard, L.A. and Loosli, J.K.1978. Animal Nutrition.Tata Me Graw Hill Pub. Company Ltd., New Delhi.613 p

Mbagwu, I.G., Adengi, H.A. and Okoye, F.C.1989. The nutritional compositions of duck weed in the Kainji lahe area and its food value to the tilapia (*sarotherodon galilaeusLinn.*)(ed. Iioba, C.).Ecological implications in the development of water bodies in Nigeria.pp. 115-124

Mendoza, R., Revol, A., Fauvel, C., Patrois, J. and Guillaume, J.C.1997. Influence of squid extracts on the triggering of secondary vitellogensis in *Penaeus vannamei. Aquaculture Nutr.*3: 55-63

Millikin, M.R., Forster, A.R., Fair, P.H. and Sick, L.V.1980. Influence of dietary protein concentration on growth, feed conversion and general metabolism of juvenile prawn (Macrobrac*hium rosenbergii). Proc.Wld. Maricult.Soc.*11: 382-391

Minamizawa, A. and Morizane, T.1970. Report on study about cultivation Technique for Fresh water shrimp. Ehime Prefecture Fisheries Laboratory, Bureau of Commercial Fisheries, office of Foreign Fisheries, U.S. Department of Interior Washington, D.C.

Mishra, B.2007.On Duckweed, The multi-use aquatic plant. *Fishing chimes* 27(5): 11-13

Mitra, G., Mukhopadhyay, P.K. and Chattopadhyay, D.N. 2005. Nutrition and feeding in Fresh water prawn (*Macrobrachium rosenbergii*) farming. Aquafeeds: Formulation and Beyond.2 (1): 17-19

Miyajima, L.S., Broderick, G.A. and Reimer, R.D.1976. Identification of the essential amino acids of the fresh water shrimp *Macrobrachium ohione. Proc.Wld.Maricult.Soc.*8: 245-250

Mohanty, S.N. and Dash, S.P.1995. Evaluation of *Azolla caroliniana* for inclusion in carp diet. *J. Aquaculture Trop.*10 (4): 343-353

Mondal, T.K. and Ray, A.K.1996. The nutritive value of *Acacia auriculiformis* leaf meal in compounded diets for rohu, *Labeo rohita*, fingerlings. The Fourth Indian Fisheries Forum. Cochin University of Sci.and Tech.Kochi. 164 p.

*Moore, B.L. and Stanley, W.R.1982. Corn silage as a feed supplement for grow-out of *Macrobrachium rosenbergii* in ponds.*J.Wld.Maricult.Soc.*13:86-94

Naik, A.T .and Murthy, H.S. 2001. Evaluation of mixed feeding schedules and varied dietary protein and lipids level in nursery and grow out farming of fresh water prawns, *Macrobrachium rosenbergii.* PhD. Thesis submitted to university of Agricultural Sciences, Bangalore, India, pp. 152

Neeraja, T.1998. Growth response of giant freshwater prawn *M.rosenbergii* juveniles to feed based on combined animal protein sources. M.F.Sc.Thesis, submitted to Kerala Agricultural University, College of Fishereies, Panangad, Cochin, and India. pp.101

New and Singholka, S.1982. Fresh water prawn farming. A manual of the culture of *Macrobracghium rosenbergii*.FAO Fish .Tech.paper, 225.FAO, Rome.116 p.

New, M.B.1976. A review of dietary studies with shrimp and prawns. *Aquaculture*. 9: 101-144

New, M.B.1990. Compounded feed stuffs for shrimp culture (eds. New, M.B., De Saram, H. and Singh, T.). Proc.Aquatech'90 Conference. Kuala Lumpur, Malaysia, 11-14 June 1990.pp.79-105

New, M.B.1996. Responsible use of aquaculture feeds. *Aquaculture Asia*.1:3-15

New, M.B.2000. History and global status of freshwater prawn farming. Freshwater prawn culture. (A farming of *M.rosenbergii*) (eds.New, M.B. and Valenti, W.C.).*Blackwell Science Ltd.*, USA, pp.1-12

Noor, J., Hossain, M.A., Bair, M.M. and Azimuddin, K.M.2000. Effects of duck weed (*Lemna minor*) as dietary fishmeal substitute for silver barb. *Bangladesh J. fisheries Research*.4 (1): 35-42

Olvera-Novoa, M.A., Martinez-Palacios, C.A., Galvan, C.E. and Chavez, S.C.1988. The use of seed of the leguminous plant *Sesbania grandiflora* as a partial replacement for fish meal in diets for tilapia (*Oreochromis mossambicus).Aquaculture*.71: 51-60

Pamungkas, W., Khasani, I. and Dewi, R.R.P.S. 2005. The effects of Vitamin C on the gonadal maturation of freshwater prawn, *Macrobrachium rosenbergii*. *Wld.Aquaculture*. Bali, Indonasia.Abstracts

Patnaik, S., Swamy, D.N., Rout, M. and Das, K.M.1991. Use of Otellia and Nymphoides leaf meal as protein source in the feed of Indian major carp. Nat.symp.On New Horizon in freshwater Aquaculture.Bhubaneswar (India), Jan 23-25.1991.pp.100-102

Patra, B.C., Maity, J., Debnath, J. and Patra, S.2002. Making aquatic weed useful: *Nymphoides cristatum* as feed for an Indian major carp *Labeo rohita (Hamilton).Aquaculture Nutr.* 8: 33-42

Paulraj, R.1993. Aquaculture feed. The marine products Export Development Authority, Kochi, India.93 p.

Pearson, D.1976. The chemical Analysis of Food. Churchill, London, 93 p.

Pezzato, L.E., Barros, M.M., Delcarrotore, C.R., Magalhaes, R.G.J. and Salaro, A.L. 1995. Growth performance of post larval Malaysian's prawn (*Macrobrachium rosenbergii*) fed at different levels of dietary protein (ed.Zimmermann, S.).Proc.6th-Rio-Grande-Meeting of Aquacult.Experts-and 3rd-South –Brazil- Meeting on Aquaculture.1995, pp. 26-33

Piedad-Pascual, F., Cruz, E.M. and Sumalangcay, Jr., A. 1990. Supplementary feeding of *Penaeus monodon* juveniles with diets containing various levels of defatted soyabean meal. *Aquaculture.* 89: 183-191

Posadas, B.C., Walters, S.C. and Long, R.D.2001. Grow out test of fresh water prawns finds effects of feed protein levels limited. *Global Aquaculture Advocate* 4(6): 28-29

Raghavan, R.and Prasad, G.2006. Towards sustainability of fresh water prawn farming-in relation to feed and feeding management. *Fishing chimes* .26(8): 36-38

Raje, P.C. and Joshi, V.P.1992. *Evaluation of different diet/diet combinations for the larvae of M.rosenbergii* (ed.Silas, E.G.). Fresh water Prawn, Kerala Agricultural University, Thrissur, India: pp. 218-222

Rath, G.S. and Dube, K.1994. Role of Zinc in promoting growth and survival of *Macrobrachium rosenbergii. J.Aquaculture.Trop.*9: 160-167

Ravishankar, A.N. and Keshavanath, P.1988. Utilization of artificial feeds by *Macrobrachium rosenbergii* (de Man). *Indian J. Animal.Sci.* 58 (7): 876-881

Ray, A.k. and Das, I.1993. Incorporation of dried aquatic weed (*salvinia cuculata*) petal in the pelleted feed for rohu, *Labeo rohita* (Lam) fingerlings. *Indian Fisheries Forum*, Pant Nagar, U.P, pp.11-14

Ray, A.K. and Das, I.1995. Evaluation of dried aquatic weed, *Pistia stratiotes*, meal as a feed stuff in pelleted feed for rohu, Labeo *rohita* fingerlings. *J. Applied Aquaculture.*5 (4): 35-44

Reed, L. and D'Abramo, L.R. 1989. A standard reference diet for crustacean nutrition research.III. Effects on weight gain and amino acid composition of whole body and tail muscle of juvenile prawns *Macrobrachium rosenbergii. J.Wld. Aquacult.Soc.* 20: 107-113

Reena, T. and Qureshi, T.A. 1996. Influence of different artificial feeds on the growth and survival of *Macrobrachium dayanum. The IV Indian Fisheries Forum Nov, 1996.School of Marine Sciences.CUSAT., Kochi, India.* Abstr: 24-28

Reigh, R.C. and Stickney, R.R.1989. Effect of purified dietary fatty acids on the fatty acid composition of fresh water shrimp, *Macrobrachium rosenbergii. Aquaculture.*77 (2): 157-174

Reyes, O.S. and Fermin, A.C.2003. Terrestrial leaf meals or freshwater aquatic fern as potential feed ingredients for farmed abalone *Halitosis asinine* (L). *Aquaculture Research* .34(8): 593-599

Richter, N., Siddhuraju, P. and Becker, K.2003.Evaluation of nutritional quality of moringa (*Moringa oleifera* Lam.) leaves as an alternative protein source for Nile tilapia (*Oreochromis niloticus L.).Aquaculture.* 217: 599-611

Roustaian,P., Kamarudin,M.S., Omar,H., Saad,C.R. and Ahmad,M.H. 2001.*Macrobrach-ium rosenbergii* larval performance, post larval production and fatty acid composition. *J.Aquaculture.Trop*.16 (3): 251-263

Routray, P. and Routray, M.D.1997. Growth potential of grass carp, *Ctenopharyngodon idella*, Val. In saline water with an aquatic weed *Potamogeton pectinatus* as feed. *Fishery technology*. 34(2): 7-10

*Sandifer, P.A. and Joseph, J.D.1976. Growth responses and fatty acid composition of juvenile prawns *Macrobrachium rosenbergii* fed on prepared ration augmented with shrimp head oil. *Aquaculture*.8: 129-139

Sandifer, P.A. and Smith, T.I.J. 1985. Freshwater prawns (eds. Hunter, J.V. and Brown, E.N.). *Crustacean and mollusk Aquaculture in the United States.* Avi publishing Co., West port. p. 63-153

Santiago, C.B., Aldaba, M.B., Laron, M.A. and Reyes, O.S.1988. Reproductive performance and growth of Nile tilapia (*Oreochromis niloticus*) broodstock fed diets containing *Leucaena leucocephala* leaf meal. *Aquacultue*.70: 53-61

Sarac, H.Z., Gravel, M., Saunders, J. and Tabrett, S.1993. Evaluation of Australian protein sources for diets of diets of the black tiger prawn (*Penaeus monodon*) by proximate analysis and essential amino acid index (eds. Carrillo, M., Dahl, L., Morale, J., Sorgeloos, P., Svennevig, N. and Wyban, J.).Abstracts of the International Conference World Aquaculture'93, May 26-28, 1993, Torramolinos, Spain.European Aquaculture Society, Special publication No.19, Oostendo, Belgium.

Shang, Y.C.1981. Aquaculture economics: Basic concepts and methods of analysis. West View Press, Colorado, 153 p.

Sheen, S. and D'Abramo, L.R.1991. Response of juvenile fresh water prawn *Macrobrachium rosenbergii* to different levels of cod liver oil/corn oil mixture in a semi purified diet.*Aquaculture*.93: 121-134

Sherief, P.M., Nair, C.M., and Malika, V.1992. *The cholesterol requirement of larval and post larval prawn (Macrobrachium rosenbergii)* (ed. Silas, E.G.).Freshwater Prawns.Proc.Nat.Symp.Freshwater prawn (*Macrobrachium spp.*), held at Faculty of Fisheries, Kochi, KAU, India, Dec.12-14, 1990. pp.213-217

Sherief, P.M.1987. Clam meat as animal protein source in the diet of Macrobrachium rosenbergii. In: Diversification of Post-harvest Technology for low cost Fish. *Society of Fisheries Technologists* (India), Cochin.pp. 327-334

Sherief, P.M.1989. Clam meat as animal protein source in the diet of fresh water prawns. (eds.Balachandran, K., Perigreen, P.A., Madhavan, P. and Surendran, P.K.).Recent Advances in Processing of Low cost Fish. *Society of Fisheries Technologies* (India), Cochin.pp. 327-334

Shewbart, K.L., Mies, W.L. and Ludwig, P.D.1972. Identification and quantitative analysis of the amino acids present in protein of the brown shrimp, *Penaeus aztecus*. *Mar.Biol.*16: 64-67

Shiav, S.Y. and Hsieh, J.F.2001. Dietary potassium requirement of juvenile grass shrimp *Penaeus monodon*. *Fisheries science*.67: 592-595

Sick, L.V., and Millikin, R.1983. Dietary and nutrient requirements for culture of the Asian prawn, *Macrobrachium rosenbergii* (eds. Mc Vey, J.P.and Moore, J.R.).CRC press Inc. Boca Ratom, Florida.pp.381-389

*Sick, L.V. and Beaty, H.1975. Development of formula feeds designed for *Macrobrachium rosenbergii* larval and juvenile shrimp. *Proc.Wld .Maricult.Soc*.6: 89-102

Siddhuraju, P and Becker, K.2001. Preliminary nutritional evaluation of mucuna seed meal (*Mucuna pruriens Varutilis*) in common carp (*Cyprinus carpio L.*): an assessment by growth performance and feed ytilization.*Aquaculture*.196: 105-123

Smith, B.R., Miller G.C. and Mead, R.W.1987. Taurine tissue concentrates and salinity effect on taurine in the fresh water prawn *Macrobrachium rosenbergii, Comp.Biochem physiol.* 87(4): 907-909

Stahl, M.S .and Aheran, G.A.1978. Amino acid studies with juvenile *Macrobrachium rosenbergii. Proc.wld. Maricult.Soc.* 9: 209-216

Stickland, J.D.H. and Parson, T.R.1972. A practical hand book of sea water analysis. *Bull.Fish.Res.Rd.Con.*167: 311-315

Sultan, S.2005. Giant freshwater prawn (GFP) farming, constaints and prospects in Up. *Fishing chimes*. 25(9): 14-18

Swamy, H.V.V. and Devaraj, K.V.1995. Growth response of common carp fry fed on three formulated feeds. *Int. j.Anim.Sci.*10 (1): 28-63

Teshima S., Koshio S., Kanazawa A. and Oshida K.1994. Essential fatty acids of the prawns *Macrobrachium rosenbergii. Proceeding of the 3rd Asian Fisheries Forum*,26-30 October 1992,Singapore (ed. Chou,L.M., Munro,A.D., Lam,T.J., Chen,T.W., Cheong,L.K.K., Ding,J.K., Hooi,K.K., Khoo,H.W., Phang,V.P.E., Shim,K.F. and Tan,C.H.) Asian Fisheries Society, Manila, Philippines. pp.705-708

Teshima, S., Koshio, S., Kanazawa, A. and Oshid, K.1992. Essential fatty acids of the prawn, *Macrobrachium rosenbergii*. 3rd Asian Fisheries Forum, October 26-30, Singapore

Tidwell, J.H., Webster, C.D. and Clark, J.A.1992. Evaluation of distillers dried grains with solubles as a protein source in diets for the fresh water prawn *Macrobrachium rosenbergii*. : Growing-Toward-The –2ist-Century. *Aquaculture*. 92: 216-217

Tidwell, J.H., Webster, C.D., Yancey, D.H. and D'Abramo, L.R.1993. Partial and total replacement of fish meal with soyabean meal and distillers by-products in diets for pond culture of the fresh water prawn (*Macrobrachium rosenbergii). Aquaculture*.118 (1-2): 119-130

Tiwari, J.B. and Sahu, N.P .1999. Possible use of Soya lecithin as a source of lipid in the post-larval diet of *Macrobrachium rosenbergii. J. Aquaculture Tropi.* 14: 37-46

Unnikrishnan, R., Nair, C.M. and Sherief, P.M.1991. Utilization of silkworm pupae in the diet of fresh water prawn, *Macrobrachium rosenbergii* (ed. Silas, E.G.).Fresh water Prawn.Proc.Nat.Symp.Freshwater prawn(*Macrobrachium spp*),held at Faculty of Fisheries ,kochi,Kerala Agricultural University,Thrissur,India,Dec12-14,1990. pp. 229-233

Vasquez, D.E., Rouse, D.B. and Rogers, W.A.1989. Growth responses of *Macrobrachium rosenbergii* at different levels of hardness. *J.Wld. Aquaculture Soc.* 20 (2): 90-92

Vasudevappa, C., Sivasankar, N. and Patil, S.A.1993. Growth of giant fresh water prawn *Macrobrachium rosenbergii* (de Man) fed on different formulated feeds. The 3rd Indian Fish Forum Proc., 11-14 October 1993, Pantnagar, pp.33-35

*Venkataramaiah, A., Cock, D.W., Biesiot, P: and Lakshmi, G.J. 1978. Nutritional value of high marsh grass and shrimp shell waste for commercial brown shrimp (*Penaeus aztecus*).Proc. *World Maricult.Soc.* 9: 217-224

Villarreal, H., Rivera, M.C. and Millan, A.1991. Effect on the substitution of shrimp meal, fish meal and Soya meal for red crab (*Pleuroncodes planipes*) meal in the growth of juvenile *Penaeus californiensis*. *Wld. Aquaculture.Soc.* PuertoRico. Abstr: 16-20

Watanabe, W.O., Ahearn, G.A. and Stahl, M.S.1976. Nutritional requirements of *Macrobrachium rosenbergii* (Unpublished Report).

*Watanabe, W.O.1975. Identification of the essential amino acids of the fresh water prawn, *Macrobrachium rosenbergii*. MSc .Thesis, Dept.Zool.University of Hawaii, pp.26

Wee, K.L.and Ng, L.T.1986. Use of cassava as an energy source in a pelleted feed for the tilapia *Oreochromis niloticus*, L. *Aquaculture Fish Manage.*17: 129-138

Xu, L.X.1994. Influence of dietary lipid source on fecundity, egg hatchability and fatty acid composition of Chinese prawn (*Penaeus chinensis)* broodstock. *Aquaculture.*119: 359-370

Zimmermann, S., Leboute, E.M., De Silva, S.M. and Penz, A.M.1991. Effects of three different protein sources in grow out feeds for freshwater prawns, *Macrobrachium rosenbergii* reared in nursery. Proceedings of 3rd Brazilian Sym.on shrimp culture, 15-20 Dec 1991.Jao Persa Paraiba, Brazil. pp. 257-287

Zimmermann, S., Leboute, E.M.and De Souza.S. 1992. The effect of four different forages in feeds for fresh water juvenile prawns, *Macrobrachium rosenbergii*. World Aquaculture'94, 14-18 January1994, New Orleans, USA.*Wld. Aquaulture Soc.*, Baton Rouge, Louisiana, USA. 196 p.

*Zimmermann, S., Leboute, E.M.and De Souza, S.M.1994. Effects of two calcium levels in diets and three calcium levels in the culture water on the growth of the freshwater prawn, *Macrobrachium rosenbergii*. World aquaculture '94, 14-18 January 1994, New Orleans, USA.*Wld.Aquaculture Soc.* Baton Roughe, Lousiana, USA.196 p.

* Not referred in original.

UTILIZATION OF WATER SPINACH *IPOMOEA AQUATICA* LEAF MEAL AS PROTEIN SOURCE IN THE FEED OF *MACROBRACHIUM ROSENBERGII* POST LARVAE

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ABSTRACT OF THESIS

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8. ABSTRACT

The present study aimed to find out the feasibility of using water spinach leaves as a low cost plant protein source and dietary evaluation of the formulated diet through growth trials employing post larvae of *Macrobrachium rosenbergii*, thus to determine the optimum level of substitution of spinach leaf meal. The experiment was done for a period of 60 days. A comparison has also been made between the diets having various levels of inclusion of water spinach leaf meal over the control diet without spinach leaf meal.

Five isonitrogenous test diets T1 to T5 were prepared with 30% crude protein. The feed ingredients used were clam meat, *Ipomoea aquatica* leaf meal, wheat bran, ground nut oil cake, tapioca flour, vitamin mineral mixture and sunflower oil. The control diet T1 was prepared by using all ingredients mentioned above, without *Ipomoea* leaf meal. The test diets T2, T3, T4 and T5 were prepared by using all ingredients mentioned above and replacing clam meat with water spinach leaf meal at inclusion levels of 12 %, 24 % 36 % and 48 % respectively.

Results showed better growth rates in prawn post larvae fed with diets (T2) containing 12% *Ipomoea* leaf meal. Among the test diets, T2 recorded highest growth rate (603.63 mg). Specific growth rate, food conversion ratio and protein efficiency ratio also showed better performance of prawn post larvae fed on test diet with 12% inclusion of spinach leaf meal. The highest SGR (3.67) and PER (2.99) were recorded in prawns fed on diet T2 and lowest SGR (2.68) and PER (1.48) were obtained with diet T5. The survival rate of post larval prawns were not found to be significantly influenced by the various test diets used and the survival ranged from 77.5% to 82.5%.