GROWTH RESPONSES OF *PENAEUS MONODON* FABRICIUS TO PELLETED FEEDS OF DIFFERENT PROTEIN SOURCES

BY

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THESIS

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Dedicated to

My beloved Father

DECLARATION

I hereby declare that this thesis entitled "GROWTH RESPONSES OF PENAEUS MONODON FABRICIUS TO PELLETED FEEDS OF DIFFERENT PROTEIN SOURCES" 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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CONTENTS

			PAGE No.
I		INTRODUCTION	1
II		REVIEW OF LITERATURE	4
	2.1	Supplementary Feeding in Prawn Culture	4
	2.2	Food and Feeding Habits of Penaeid Prawns	7
	2.3	Nutrient Requirements of Prawns	9
	2.3.1	Proteins	10
	2.3.1.1	Amino Acids	11
	2.3.2	Lipids	12
	2.3.3	Carbohydrates	13
	2.3.4	Vitamins and Minerals	14
	2.4.	Protein Sources used in Supplementary Feeds	16
	2.4.1	Plant Protein Sources	16
	2.4.2	Animal Protein Sources	17
	2.4.3	Mixed Protein Sources	20
	2.4.4	Single Cell Protein Sources	21
	2.5	Oxygen Consumption, Ammonia Excretion and O : N Ratios	23
	2.6	Ration Size and Feeding Frequency	24
	2.7	Water Stability of the Artificial Feeds	26
	2.8	Keeping Quality of Artificial Diets	28
III		MATERIALS AND METHODS	31
	3.1	Preparation of the Feed	31
	3.1.1	Feed Ingredients	31
	3.1.2	Proximate Composition of Feed Ingredients	32

	3.1.3	Formulation and Processing of Test Diets	32
	3.1.4	Proximate Composition of Formulated Feeds	33
	3.2	Sinking Rate of Pelleted Feeds	35
	3.3	Water Stability of Pelleted Feeds	35
	3.4	Total Heterotrophic Bacteria and Fungi	35
	3.5	Keeping Quality of Pelleted Feeds	36
	3.6	Experimental Animals	36
	3.7	Experimental Tanks and Water Management	36
	3.8	Study to Evaluate the Protein Source	37
	3.8.1	Determination of Digestability Coefficient	38
	3.8.2	Monitoring of Physico-Chemical Parameters of Water	38
	3.8.3	Recording of the Data	38
	3.8.3.1	Survival Rate	38
	3.8.3.2	Growth	39
	3.8.3.3	Specific Growth Rate	39
	3.8.3.4	Food Conversion Ratio	39
	3.8.3.5	Protein Efficiency Ratio	39
	3.8.3.6	Protein Digestability Coefficient	40
	3.8.4	Biochemical Composition of Carcass	40
	3.8.5	Estimation of Oxygen : Nitrogen Ratios	40
	3.9	Study to Determine the Optimum Food Ration	41
	3.10	Study to Determine Optimum Feeding Frequency	41
	3.11	Statistical Analysis	42
IV		RESULTS	43
	4.1	Proximate Composition of Feed Ingredients and Formulated Feeds	43

	4.1.1	Feed Ingredients	43
	4.1.2	Formulated Pelleted Feeds	45
	4.1.2.1	Initial Analysis	45
	4.1.2.2	Analysis After Storage	45
	4.2	Sinking Rate of Pellets	47
	4.3	Water Stability	47
	4.4	Total Heterotrophic Bacteria and Fungi	49
	4.5	Evaluation of Relative Efficiencies of Natural Protein Sources	49
	4.5.1	Physico-chemical Parameters	49
	4.5.1.1	Temperature	49
	4.5.1.2	pH	49
	4.5.1.3	Dissolved Oxygen	55
	4.5.2	Growth in Relation to Protein Source	55
	4.5.2.1	Survival Percentage	55
	4.5.2.2	Growth	55
	4.5.2.3	Specific Growth Rate	58
	4.5.2.4	Food Conversion Ratio	58
	4.5.2.5	Protein Efficiency Ratio	64
	4.5.2.6	Protein Digestability	64
	4.5.3	Biochemical Composition of the Prawn	72
	4.5.4	Oxygen Consumption, Ammonia Excretion and O : N Ratios	79
	4.6	Study on Optimum Food Ration	84
	4.7	Study on Optimum Feeding Frequency	89
V		DISCUSSION	94
	5.1	Keeping Quality of Formulated Feeds	94
	5.1.1	Moisture	94

	5.1.2	Proteins	95
	5.1.3	Fat	95
	5.1.4	Ash, Crude Fibre and Carbohydrate	96
	5.1.5	Total Heterotrophic Bacteria and Fungi	96
	5.2	Water Stability of Pelleted Feeds	96
	5.3	Evaluation of Efficiencies of Natural Protein Sources	98
	5.3.1	Feeding Experiment	98
	5.3.1.1	Growth Studies	98
	5.3.1.2	Food Conversion Ratio	102
	5.3.1.3	Protein Efficiency Ratio	103
	5.3.1.4	Protein Distability Coefficient	104
	5.3.2	Biochemical Composition of the Experimental Prawns	104
	5.3.3	Oxygen Consumption, Ammonia Excretion and O : N Ratios	107
	5.4	Study on Optimum Food Ration	108
	5.5	Study on Optimum Feeding Frequency	109
VI		SUMMARY	111
VII		REFERENCES	114
VIII		ABSTRACT	

LIST OF TABLES

PAGE No.

1. Proportion of different ingredients used in the formulated feeds 34

2. Proximate composition of the ingredients used in feed formulation 44

3. Proximate composition of formulated feeds 46

4. Effect of storage on the keeping quality of pelleted feeds 48

5. Temperature range of the experimental tanks during the study of protein source evaluation 50

6. Fluctuations of pH in the experimental tanks during the study of protein source evaluation 51

7. Variations in dissolved oxygen content of the experimental tanks during the study of protein source evaluation 53

8. Average weight and length attained by P. monodon juveniles fed on diets based on different protein sources for a period of 43 days 59

9. Specific growth rate of P. monodon juveniles fed on diets based on different protein sources 61

10. Food conversion ratio of P. monodon juveniles fed on diets based on different protein sources 65

11. Protein efficiency ratio of P. monodon juveniles fed on diets based on different protein sources 67

12. Protein digestability coefficient of the formulated feeds fed to P. monodon juveniles for a period of 20 days 69

13. Biochemical composition of P. monodon fed with diets based on different protein sources 74

14. Oxygen consumption, ammonia excretion rate and O : N ratios of P. monodon juveniles fed with diets based on different protein sources 80

15. Effect of food ration on food consumption and growth of P. monodon juveniles 85

16. Water quality parameters of the experimental tanks recorded during the study on effect of food ration 86

17. Effect of feeding frequency on food consumption growth and survival of P. monodon juveniles 90

18. Water quality parameters of the experimental tanks recorded during the feeding frequency study 91

ANOVA TABLES

1. Analysis of variance of the data on survival of P. monodon juveniles fed on diets based on different protein sources 57

2. Analysis of variance of the data on the gain in length and weight of P. monodon juveniles fed on diets based on different protein sources 60

3. Analysis of variance of the data on specific growth rate of P. monodon juveniles fed on diets based on different protein sources 62

4. Analysis of variance of the data on food conversion ratio of P. monodon juveniles fed on diets based on different protein sources 66

5. Analysis of variance of the data on protein efficiency ratio of P. monodon juveniles fed on diets based on different protein sources 68

6. Analysis of variance of the data on protein digestability coefficient of formulated feeds fed to P. monodon juveniles 70

7. Analysis of variance of the data on biochemical composition of P. monodon fed with diets based on different protein sources 76

8. Analysis of variance of the data on oxygen consumption, ammonia excretion and O : N ratios of P. monodon juveniles fed with diets based on different protein sources 82

9. Analysis of variance of the data on gain in length and weight of P. monodon juveniles obtained during the study on the effect of food ration 87

10. Analysis of variance of the data on gain in length of P. monodon juveniles obtained during the study on the effect of feeding frequency 92

LIST OF FIGURES

PAGE No.

1. Fluctuations of pH in the experimental tanks during the study of protein source evaluation 52
2. Variation in dissolved oxygen content (ppm) of the experimental tanks during the study of protein some evaluation 54
3. Percentage survival of *P. monodon* juveniles fed on diets based on different protein sources 56
4. Food conversion ratio of *P. monodon* juveniles fed on diets based on different protein sources 56
5. Average weight (g) and length (cm) attained by *P. monodon* juveniles fed on diets based on different protein sources 63
6. Specific growth rate of *P. monodon* juveniles fed on diets based on different protein sources 63
7. Protein efficiency ratio of *P. monodon* juveniles fed on diets based on different protein sources 71
8. Protein digestability coefficient of formulated feeds fed to *P. monodon* juveniles 71
9. Biochemical composition of *P. monodon* juveniles fed with diets based on different protein sources 75
10. Oxygen consumption and ammonia excretion rate of *P. monodon* juveniles fed with diets based on different protein sources 81
11. O : N ratio of *P. monodon* juveniles fed with diets based on different protein sources 81
12. Effect of food ration on specific growth rate of *P. monodon* juveniles 88
13. Effect of feeding frequency on food consumption and specific growth rate of *P. monodon* juveniles 93

I. INTRODUCTION

I INTRODUCTION

The tiger prawn Penaeus monodon Fabricius is the most extensively cultured crustacean in the countries of Southeast Asia, owing to its fast growth rate, capacity for adaptation to various culture systems and efficient response to compounded feed (AQUACOP, 1976; Alava and Lim, 1983). It is an ideal species for large scale farming in estuarine and brackishwater ecosystems.

Successful rearing of penaeid prawns necessitates the development of a nutritionally balanced artificial diet, through which higher stocking densities and shorter culture periods become possible. It has been observed that the cost of supplementary feed accounts to nearly 40-60% of the total operational cost in intensive farming operations (Anon, 1983). Formulation of a balanced feed containing low cost protein ingredients can bring down the cost of supplementary feed to a great extent. Hence development of an economically viable and biologically efficient diet for prawns is highly warranted. Specific nutritional requirements of the candidate species should be taken into consideration while preparation of supplementary feeds, and design of diet aimed at optimising growth and survival under large scale farming operations.

Determination of nutrient requirements has been aimed at replacing live food with formulated artificial diets. Unlike natural food, artificial diets are not subjected to seasonal variations in supply and can be manufactured under strict quality control. As shrimp feeds become better formulated in accordance with the research findings of balanced nutrient requirements and better texturised to improve acceptability, they are likely to show improved utilization. As in fishes crustaceans also show preferential use of proteins over carbohydrates. Because of this, and the fact that proteins in a compounded feed forms the most expensive item, studies on evaluation of cheaper protein sources as ingredients for supplementary feed have received much attention in the recent years. In this regard, a variety of natural feed sources are currently being utilised the world over. As against the highly nutritious practical feeds used in the developed countries, which make use of high quality feed ingredients, developing countries rely on mostly nutritionally poor quality raw materials for feed formulation. But increased competition from animal husbandry has led to shortage of conventional feed ingredients which in turn has paved the way for development of a new set of unconventional protein sources, the most important of which is single cell protein source. Their importance is not only because of their high nutritive value, but also because they can be produced on a large scale from waste products which otherwise cause environmental hazards. Though a number of studies have been made in the past for incorporation of single cell protein in the diet of fishes, studies in this direction are limited in the case of prawns.

In India, detailed research on penaeid prawn nutrition was taken up only recently when commercial culture of prawns gained momentum. One of the areas of active research has been the feed formulation technology for the penaeids and suitable feeds are being developed both for larvae in hatcheries and the juveniles in grow-out systems. The types of feeds used at present in various grow-out systems include conventional feeds of different oil cakes and brans, cattle feeds available in the market, trash fishes, clam meat, meat offals, different types of pelleted feeds etc. Many of the feeds are inadequate to meet the nutritional requirements of the prawns. Moreover the exorbitant cost of the pelleted feed is another hindrance for its large scale use in prawn farming. Hence formulation of a low cost, nutritionally balanced prawn feed containing adequate levels of protein is essential.

In the present study an attempt has been made to evaluate the efficiency and keeping quality of pelleted feeds formulated from different protein sources such as clam meal, prawn meal, slaughter house waste meal, soyabean meal, and single cell protein (marine yeast) for P. monodon juveniles. The optimum food ration and feeding frequency of the selected diet were also studied in view of their practical utility in farming operation.

II. REVIEW OF LITERATURE

II REVIEW OF LITERATURE

2.1 Supplementary feeding in Prawn Culture

In the traditional prawn and fish culture practices in the brackishwater regions, followed in India and some of the Southeast Asian countries, supplementary feeding of the population is not practised and the prawn and fish grow by feeding on the natural food available in the ponds. According to Hepher and Ghervinski (1965), intensification of culture practices and large scale stocking of fish and prawn may lead to depletion of natural food available in the environment which in turn may affect their growth. Supplementary feeding is the only satisfactory solution to this problem. The most important ingredients used in supplementary feeds are clam, mussel, trash fish, animal flesh, silk worm pupae, bone meal, liver, blood meal, eggs, oil cakes obtained from ground nut, coconut, gingelly, mustard and soyabean, rice bran, wheat bran, and several kinds of grains either singly or in combination (Spinelli et al., 1978; Hepher and Pruginin, 1981).

With the rapid progress of semi-intensive and intensive systems of culture, formulation and development of practical feeds meeting the nutritional requirements of candidate species for obtaining optimum growth and survival during the different phases of its growth were evolved. Accordingly, during the past two decades considerable advancements have been made in the various aspects of feed technology and several types of compounded feeds have been developed and are being used for the culture of prawns in different countries of the world (Japan - Shigueno, 1984; Philippines - SEAFDEC, 1981 & 1983; Liu and Mancebo, 1983; Apud, 1984; Pascual, 1984; Tibbu et al., 1984; Taiwan - Liao, 1981; Chiang and Liao, 1985; Tahiti - AQUACOP, 1984 a & b; United States - Parker and Fred, 1974; Elam and Green, 1974; Huang et al., 9184; Lee and Shlesser, 1984; Latin America - Escobar, 1984; Scura, 1985).

One of the earliest published work of compounded rations for prawn was that of Kanazawa et al. (1970), who designed a diet for P. japonicus. Deshimaru and Shigueno (1972) used a diet made up of several ingredients such as squid meal, fish meal, mysid meal, shrimp meal, yeast, soyabean meal, active sludge, casein and gelatin for rearing the prawn P. japonicus. They found that the diet having amino acid composition similar to the amino acid composition of prawn gave good results. Hence they prepared diets with amino acid composition similar to that of P. japonicus by using a mixture of animal and plant protein sources and established that prawns could be successfully cultured on such artificial diets. Elam and Green (1974) conducted feeding experiments with different formulated feeds on P. setiferus and obtained food conversion ratios ranging from 1.8 to 2.3. Colvin (1976) studied the growth, digestability and food conversion ratios of some diets formulated with fish meal and shrimp meal in P. indicus and reported a growth rate of about 0.44 mg/day and lowest food conversion ratio of 2.72. Feeding trials conducted on P. monodon cultured in cages using a practical feed in Philippines (SEAFDEC, 1981) had shown a food conversion ratio of 4.8. In semi-intensive culture experiments with the same species (SEAFDEC, 1983) commercial prawn feed with 45% protein and an experimental feed with 35% protein produced food conversion ratios of 3.4 to 4.6 and 6.1 respectively.

In India also attempts have been made to develop practical feeds with locally available materials by Alikunhi _et al._ (1980); Ahamad Ali and Sivadas (1983) and Mohamed _et al._ (1983) to rear prawn larvae and post-larvae. Suitable artificial feeds were also formulated by a number of workers for feeding the prawns in the grow-out systems. Two sets of supplementary feeds were used in 1978 for rearing _P. monodon_ post larvae at Kakdwip research centre of CIFRI.* The first set consisted of soyabean flour, Brewer's yeast, maize powder, wheat bran, calcium phosphate, vitamin and algin and the second set of goat offal, yeast algal power, wheat flour and terramycin. It was reported that survival of post larvae fed with above diets ranged from 57% to 90% (Anon, 1978). Specific studies on feeds and nutrition of _Penaeus monodon_ were also carried out at Kakdwip fish farm using different feed formulations by Rajyalakshmi _et al._ (1979). Protein percentages in the feeds were maintained at 20 to 36% with both animal and plant protein sources. Feed containing squid meal and soyabean meal gave the best growth and food conversion. Goswami and Goswami (1979) evaluated the growth performance and conversion efficiency in penaeid prawns with feeds prepared from slaughter house products and poultry processing byproducts.

Raman _et al._ (1982) conducted three sets of yard experiments to determine the effects of supplementary feeds prepared from cheap and locally available materials on growth and survival of _P. indicus._ A combination of fish meal, rice bran and tapioca in two proportions of 1:1:1 and 2:2:1 respectively proved to be the best among the feeds. Ahamad Ali (1982a) conducted experiments in juveniles of _P. indicus_ and reported that feed

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* Central Inland Fisheries Research Institute

containing clam meal powder gave a food conversion of 1.46, mantis shrimp of 1.71 and prawn waste meal of 2.27. Mohammed Sultan et al. (1982) formulated feeds with frog flesh and reported a food conversion ratio 3.01 to 4.96 for P. indicus and 5.87 to 8.21 for P. monodon. Although the above endeavours have provided certain informations on the use of compounded feeds, the results and production rates obtained have not been consistent.

2.2 Food and feeding habits of Penaeid Prawns

Investigations on food and feeding habits of prawns have helped to develop suitable supplementary feeds and feeding practices. Prawns inhabit the benthic region and feed on live animals and vegetable matter as well as dead organic matter. According to Chopra (1939) the prawns eat all types of food, living or dead that comes in its way. Gopalakrishnan (1952) observed that crustaceans and vegetable matter formed bulk of gut contents of P. indicus. Panikkar (1952) has reported that food of young penaeids consisted of algae, minute organisms and organic detritus. Panikkar and Menon (1956) also found that P. indicus, M. dobsoni, Parapenaeopsis stylifera were mainly detritus feeders, although small amounts of animal remains, sand and mud were encountered in their gut contents. Hall (1962) considered penaeids to be omnivores, with P. monodon in particular preferring crustaceans, vegetable matter, polychaetes, molluscs, fish and insects. Subrahmanyam (1963) concluded that P. monodon is carnivorous in habit. This view is supported by Thomas (1972) who studied the feeding habits of P. monodon from Korapuzha estuary and has reported that the contained mainly crustaceans, molluscs, polychaetes and fishes. Kuttyamma (1973) observed that debris composed of mud and organic matter constituted the main portion of the stomach contents in P. monodon, while crustaceans ranked next in quantity. Mohanty (1975) reported that the stomach contents of P. monodon include 29.78% of food of animal origin while Rao (1967) estimated it to be 40.77%. Marte (1980) reported that the food of P. monodon consisted of crustacea (small crabs, and shrimps) and molluscs making up 85% of ingested food, the remaining 15% consisted of fish, polychaetes, ophiuroids, debris, sand and silt. Su and Liao (1986) also observed more or less similar items in the gut contents of P. monodon. All these findings suggest that P. monodon is more of a carnivore with preference to macro-invertebrates rather than a scavenger or debris feeder.

Feeding intensity of P. monodon during different months in a year revealed that the species feeds more intensively during February to May and moderately during June to September (Mohanty, 1975). According to Marte (1980) P. monodon seems to have increased feeding activity during ebb tide. Significant monthly variations in feeding activity were shown by the wild prawn caught from Mikato area (Marte, 1982). The species feeds by seizing the food with its pincers and pushing it to mouth (Villadolid and Villaluz, 1951). If the feed stuff in the pellet is not homogenous enough, larger particles are spit out. If the whole pellet is small enough for their mouth, it is consumed (Pascual, 1988). Cannibalism is caused by factors such as insufficient food, crowding, poor quality of feed or nutritionally inadequate ration. Although P. monodon has been found to feed continuously all day, it seems to consume more food at night than during the day (Pascual, 1988). Apud et al. (1980) have observed that P. monodon in ponds eat at anytime of the day but prefer to feed at bottom when there is light. The prawns move around the perimeter of the pond in late afternoon and evening, and hence suggestions are to give more feed during this time (Pascual, 1988).

2.3 Nutrient requirements of Prawns

The results of various investigations carried out on the nutrition and nutritional requirements of prawns have been reviewed by New (1976), Castell et al. (1981); Claybrook (1983) and Dall and Moriarty (1983). New (1980) compiled a bibliography of prawn and shrimp nutrition, and Pruder et al. (1983) reviewed works on penaeid nutrition. Kanazawa (1984) also reviewed the recent advances made in peanaeid nutrition.

Nutrition in crustaceans as such is complicated by a number of abiotic and biotic factors which have tremendous influence on growth and utilization of food. Abiotic factors such as temperature, pH, salinity, dissolved oxygen, depth, light etc, have been found to directly affect growth and food utilization of crustaceans (Subrahmanyam, 1962; Venketaramiah et al., 1974; Vernberg, 1983). Amongst the biotic factors, moulting which forms an important event in the life cycle has considerable influence on the growth and feed utilization of prawns. It has been established that each moulting in crustacea results in considerable energy loss ie about 7.3% per moult in Macrobrachium rosenbergii and potentially and average rate of 0.81% per day is lost (Nelson et al., 1977). It amounts to a large quantum of energy since at this stage the prawn moults every 8 to 10 days or even earlier (Stern, 1976).

2.3.1 Proteins

Among the nutrients, protein is the most important, as it forms the major growth factor in animal tissues. So greater emphasis was given for understanding the protein requirement and for determining the optimum protein levels in the diet of different species (Kanazawa et al. (1971a); Deshimaru and Shigueno (1972); Balazs et al. (1973); Forster and Beard (1973); Deshimaru and Kuroki (1975a); Colvin (1976); Khannapa (1979); Kanazawa et al. (1981). These results indicate that protein requirement ranging from 15 to 80% is needed for different species of penaeid prawns. Deshimaru and Yone (1978a) have pointed out that P. japonicus requires 52 to 57% protein for optimum growth and food conversion efficiency. Kanazawa et al. (1981) have demonstrated that Metapenaeus monoceros showed best growth with a diet containing 55% casein. Colvin (1976) has reported, the optimum protein level for P. indicus as 43%. In the case of Penaeus monodon it was shown to be 46% by Lee (1971); 40% by AQUACOP (1977); Khannapa (1977); and Alava and Lim (1983); 35% by Bages and Sloane (1981); and 50% by Millamena et al (1986). Bautista (1986) in a recent study on P. monodon has shown that 40 to 50% protein gave best growth and survival in the presence of 20% carbohydrate and 5 to 10% lipid. Nezaki et al. (1986a) found that 55% protein with 15% carbohydrate in grow-out diets gave the best growth for P. monodon. However according to them, when the carbohydrate content is increased to 25%, a 45% protein diet can give results comparable to those diets containing 55% protein.

In Penaeus aztecus the protein requirement was reported to be 23 to 31% (Shewbart et al., 1973; Venketaramiah et al., 1975). In the case of *P. setiferus* it was 28 to 32% (Andrews *et al.*, 1972). Colvin and Brand (1977) have shown the protein requirement for *P. californiensis* to be 31% and for *P. vannamei* to be 30%. For *P. merguiensis* it was shown to be 50% by AQUACOP, (1978) and 34 to 42% by Sedgwick (1979). The variation in protein requirement can be attributed to various factors such as differences in biological value of protein sources which again depends on the amino acid composition of protein (Harper, 1981; Kies, 1981); composition of other dietary components, viz. fat and carbohydrate ratio (Andrews *et al.*, 1972; Teshima and Kanazawa, 1984) and amount of organic salts present in the diet (Deshimaru and Kuroki, (1974); New, 1976). Variation in environmental factors also contributes to a certain extent to the difference in protein requirement.

2.3.1.1 Amino acids

Amino acids being the building blocks of protein, their profile in the protein source greatly determines the efficacy of its utilization. Shewbart *et al.* (1972) investigated the amino acid requirement of *P. aztecus*. Kanazawa and Teshima (1981) have elucidated by tracer techniques using radioactive acetate, that *P. japonicus* requires ten amino acids ie, argenine, methionine, valine, threonine, isoleucine, leucine, lysine, histidine, phenyl alanine and tryptophan all of which are also essential amino acids for various fishes. The requirements of these essential amino acids have also been demonstrated for other penaeids such as *P. monodon* (Coloso and Cruz, 1980) and *P. aztecus* (Shewbart *et al.*, 1972). It was also shown by Deshimaru and Kuroki, (1975b) that diets containing only aminoacids instead of protein brought about a very poor growth and high mortality in feeding trials of *P. japonicus.* Wilson (1984), has shown that, the closer the essential amino acid pattern of the diet to that of the cultured species, the more effective is the diet for its growth.

2.3.2 Lipids

Lipids are important for their high energy value, polyunsaturated fatty acids (PUFA) phospholipid content and vitamins (A, D, E & K). Prawns have specific qualitative requirement of lipids rather than their quantity. A lipid level of below 10% was found to be adequate in the prawn diets (Andrews *et al.*, 1972; Forster and Beard, 1973). Employing radioisotope tracer technique, Kanazawa and Teshima (1977) and Kanazawa *et al.* (1977) have shown that prawns are not capable of synthesizing polyunsaturated fatty acids such as linoleic acid (18 : 2W6), linolenic acid (18 : 3W3), eicosapentaenoic acid (20 : 5W3) and decosa hexaenoic acid (22 : 6W3). These fatty acids are essential for prawns and should be supplied in their diet. In fact Kanazawa (1984) has demonstrated, that the diets containing the above fatty acids produced faster growth rate in *P. japonicus.* Mendoza (1982) found that a diet containing 11.7% lipid gave maximum growth, efficient feed conversion, protein efficiency ratio and good survival in *P. monodon* juveniles. Catacutan and Kanazawa (1985) showed that the best growth of *P. monodon* juveniles was attained, with lipid source containing high amounts of highly unsaturated fatty acids of the **W**3 Series in semipurified diets, the quantity being 10 to 11% of the diet. Bautista (1986) also showed that a 10% lipid content in the diet of *P. monodon* was effective in assuring good growth and survival, as long as protein content was between 40 and 45% and carbohydrate was 25%.

Other types of lipids needed by the prawns are cholesterol (Nalzaro, 1982) and phospholipid (Pascual, 1986). Feeding experiments using artificial diets have shown that P. japonicus requires 0.5% cholesterol for maximum growth and survival (Kanazawa et al., 1971b). But Nalzaro (1982) reported that, a total cholesterol level of 1% is required in P. monodon juveniles for maximum growth, high food conversion, high protein efficiency, high survival rate and maintenance of a constant level of body cholesterol. Ubal et al. (1986) indicated that a level of 0.6% to 1.0% cholesterol in a semi-purified diet containing 6% refined cuttle fish liver oil and soya oil in 3:1 ratio gave good weight gain and feed efficiency in P. monodon juveniles.

Lecithin which contains 62% phospholipids; phosphatidyl choline, phosphatidyl inositol, and phosphatidyl ethanolamine, has been found necessary in prawn grow-outs at around 3% in the diet (Pascual, 1986). Nazaki et al. (1986b), showed that soyabean lecithin levels at around 4% gave the best results, compared to those fed greater than 4.6% soyabean lecithin levels. Percentage weight gains and feed efficiency increased significantly in P. monodon juveniles and decreased with increasing lecithin levels beyond 4%, Millamena et al. (1986) reported a level of 4% lecithin was necessary for brood stock diets.

2.3.3 Carbohydrates

Carbohydrates in prawn diets are not only useful for their energy value and protein sparing function but also for their binding properties.

The nutritive value of carbohydrates in the diet of prawns was investigated by a number of workers (Cowey and Forster, 1971; Forster and Gabbot, 1971; Sick and Andrews, 1973; Ahamad Ali, 1982b; Alava and Pascual, 1987), who found that penaeid prawns generally utilize disaccharides and polysaccharides better than monosaccharides, and a carbohydrate level of 5 to 40% is needed in their diet. The carbohydrates that have been studied in prawn diets include, sucrose, dextrin, maltose, molasses, trehalose, glucose etc (Pascual et al., 1983; Alava and Pascual, 1987). Alava and Pascual (1987) showed that sucrose and trehalose as better sugars than glucose, for P. monodon juveniles. Addition of 20% glucose in the diet for P. aztecus showed reduced growth rate (Andrews et al., 1972). It was postulated that dietary glucose was rapidly but inefficiently utilised for energy metabolism whereas the glucose from digested polysaccharides was absorbed more effectively (Andrews and Sick, 1972). Different levels and types of dietary carbohydrates were found to affect body composition. The 40% glucose diet produced lower lipid levels, than 10% starch or glucose, but not significantly different levels from the 40% starch diet. Animals fed on 40% glucose had lower protein levels than those fed with 40% starch or 10% glucose. High levels of dietary starch have been used for their binding qualities in several experimental and commercial diets (Balazs et al., 1973; Sick and Harris, 1974).

2.3.4 Vitamins and Minerals

Vitamins and minerals are important for regulating body processes in prawns. Requirements of vitamins and minerals in the diet of prawns were investigated by several workers (Fisher, 1960; Kanazawa et al., 1970; Kitayama et al., 1971; Forster and Gabbot, 1971; Forster and Beard, 1973; Balazs et al., 1974). They found that B vitamins are necessary for proper utilization of protein, carbohydrates and fats, while vitamin A and C are important in building resistance to infection, Vitamin D together with minerals, calcium, and phosphorus is necessary for the formation of the exoskeleton or shell. All these materials although needed in minute amounts are necessary for the proper utilization of food by the prawns. Hence these nutrients should also be included in complete diets for prawns in their proper amounts. Kitabayashi et al. (1971b) observed accelarated growth in P. japonicus fed with diets containing Vitamin C. Deshimaru and Kuroki (1976) have shown that juveniles of P. japonicus require 300 to 1000 mg of ascorbic acid, 60 mg of choline, 200 to 400 mg of inositol, 6 to 12 mg of thiamine, and 12 mg of pyridoxine per 100 gms diet.

The importance of carotenoids in prawn diets for the pigmentation had been demonstrated by Joseph and Williams (1975), and Sandifer and Joseph (1976).

In the case of minerals, the requirement of calcium and phosphorus in the diet of P. japonicus was studied by Deshimaru and Yone, 1978b; Deshimaru et al., 1978) and of P. aztecus by Colvin and Brand (1977). The best growth rates of P. japonicus were achieved with diets where levels of 1.04% phosphorus and 1.24% calcium were added (Kitabayashi et al., 1971a). Inclusion of mineral premix at the rate of five per cent, in a casein based diet for P. aztecus gave 18% increase in biomass (Sick et al., 1972). Kanazawa et al. (1984) reported the requirement of calcium, phosphorous, magnesium potassium, copper, iron and manganese in the diet of P. japonicus. Bautista (1986) pointed out the importance of calcium phosphorus rations in the diets of P. monodon and found a 1:1 ratio to be effective in hardening the exoskeleton and preventing soft shell disease.

2.4 Protein sources used in supplementary feeds

2.4.1 Plant protein sources

Feeds stuffs of vegetable origin are as a whole lower in protein content when compared with those of animal origin. In addition, the presence of high amounts of carbohydrates, fibre and other organic molecules such as glucosides, phytates, and cyclopropanes in these sources, present the nutritionist with problems that are generally not encountered with sources of animal origin (Spinelli et al., 1979).

Most plant proteins have been shown to yield poor growth rates in prawns when used individually excepting a few like soyabean meal, (Kanazawa et al., 1970; Sick and Andrews, 1973; Millikin et al., 1980; Maguire and Hume, 1982), wheat gluten (Forster and Gabbot, 1971; Deshimaru and Shigueno, 1972) and peanut meal (Lee, 1970; Sick et al., 1972; Forster and Beard, 1973; Balazs et al., 1973). The improved growth rates produced by some of the plant protein sources have been attributed to their higher polysaccharides contents compared to monosaccharides (Forster and Gabbot, 1971); Kitabhayashi et al., 1971b; Andrews et al., 1972; Sick and Andrews, 1973).

Kanazawa et al. (1970) reported that soyabean meal is the best protein source among plant proteins for *P. japonicus*. Deshimaru and Shigueno (1972) observed that the aminoacid profile of the prawns is similar to that of soyabean to a large degree which may be the reason for higher growth rate in prawns fed with soyabean meal. But when compared to animal sources it is on the lower side. Sick and Andrews (1973) also made the same observation in *P. duorarum*. Sure (1948) found relatively higher levels of lysine and threonine in soyabean meal than groundnut oil cake. The above two amino acids have been shown to be essential for growth in prawns by Cowey and Forster, 1971 and Shewbart *et al.*, 1972. Besides, groundnut oil cake and coconut oil cake contain relatively higher levels of unconsumable carbohydrates such as cellulose, hemicellulose, gums and pectin in their cell walls compared to soyabean meal, which are not acted upon by the digestive enzymes and excreted mainly through faeces and this results in reduced growth rate in prawns (Swaminathan, 1967). However, Gopal (1986) has reported higher growth and survival for *P. indicus* juveniles fed on groundnut oil cake based feed.

Viola *et al.* (1981) reported that soyabean meal can successfully replace fish meal in the diet of fish. High growth rate and conversion efficiency were reported when mangrove foliage was incorporated in the diet of *P. indicus* juveniles (Sambasivan and Krishna Murthy, 1986).

2.4.2 Animal protein sources

Animal protein sources are found to be better for feed formulation of prawns than plant protein sources. Among the animal protein sources, proteins of marine origin are found to be superior than those of freshwater or terrestrial origin, not only due to their amino acid profile but also due to the better composition of their unsaturated fatty acids essential for prawns (Cowey and Sargent, 1972) as well as higher ash content (Boghen and Castell, 1981).

One of the earliest artificial diet of prawn *P. japonicus* was derived from that of a silk worm pupae, chinook salmon and brine shrimp (Kanazawa *et al.*, 1970). Later in 1972, Deshimaru and Shigueno used a diet made up of several ingredients such as squid meal, fish meal, mysid shrimp meal, yeast, active sludge, casein and gelatin for the same species. Fresh clam meat was conventionally used as feed for prawns by Kanazawa *et al.* (1970) who reported that fresh short necked clam *Tapes philippinarum* gave superior growth rates compared to compounded diet in *P. japonicus*. Similar results were also obtained by Forster and Beard (1973) in *Palaemon serratus* with fresh mussel meat. But Vellegas (1978) showed that the growth and survival of *P. monodon* larvae fed with *Tapes* was only next to that of compounded diet. Ahamad Ali (1982a) used fresh clam meat of *Sunetta scripta* as control diet for *P. indicus* while evaluating the suitability of various protein sources, but found poor performance of fresh clam meat and observed heavy mortality.

Prawn meal has also been found to be a suitable protein source for the formulation of feed for penaeid prawns. Sick and Andrews (1973) obtained higher growth and survival in *P. duorarum* fed on diets with shrimp meal. Forster and Beard (1973) also indicated that shrimp meal is nutritionally superior to fish meal in the diet of *Palaemon serratus*. Superiority of prawn meal may be due to its amino acid profile, which may satisfy the nutritional demand of the juvenile prawn. As stated earlier, diets with an amino acid profile similar to shrimp and rich in basic amino acids could produce relatively higher growth rate (Deshimaru and Shigueno, 1972). Forster (1976), reported that prawn waste contain several essential amino acids which induce high growth rate in prawns. Prawn head oil was found to contain polyunsaturated fatty acids essential for crustaceans (Joseph and Meyers, 1975). Joseph and Williams (1975) considered it as potential additive in the feeds of marine animals. Sandifer and Joseph (1976) found that waste shrimp heads of *P. setiferus* to be a good source of fatty acid and pigments in the diet of fresh water prawn *Macrobrachium rosenbergii.* Pascual and Destajo (1978) reported that shrimp head meal is one of the most promising protein source in the diet of *P. monodon.* However the same authors observed slow growth and survival of post-larvae of *P. monodon* fed with diet prepared exclusively from shrimp head meal and concluded that a diet prepared from shrimp head alone is not sufficient to produce good growth and survival. Prawn waste was used in feeds for culturing prawns in grow-out ponds (Mohamed *et al.* 1983; Ahamad Ali and Sivadas, 1983; Ahamad Ali and Mohamed, 1985). According to Vaitheswaran and Ahamad Ali (1986) growth promoting effect of shrimp head meal may be due to the presence of higher levels of calcium, phosphorus and chitin present in it.

Fish meal has also been reported to be a good source of protein with essential amino acids and has high biological value. Composition and quality of fish meal varies according to the raw materials used for preparing it. Deshimaru and Shigueno (1972) observed that diets prepared with fish meal were inferior to short necked clam for *P. japonicus.* They also found that the aminoacid composition of fish meal is not similar to that of the prawn P. japonicus. Colvin (1976) and Ahamad Ali (1982a) obtained poor results with fish meal based diets in P. indicus. Colvin (1976) suggested that relative deficiency of amino acids tyrosine and phenyl alanine in the fish meal may be the reason for the relatively poor performance of the prawn fed on fish meal based diets.

Shigueno et al. (1972) reported higher growth rates in P. japonicus fed with squid meal as protein source. In many studies squid meal was used as protein source because of its high content of argenine (Deshimaru and Shigueno, 1972; Kittaka, 1976; Fenucci and Zein-Eldin, 1976). Diets based on blood meal produced poor growth rates in P. aztecus (Colvin and Brand, 1977). However higher growth rates and conversion efficiency were obtained for prawn when slaughter house products (beef, liver) and poultry byproducts were incorporated in the diet (Goswami and Goswami, 1979).

2.4.3 Mixed protein sources

A mixture of two or more protein sources invariably show better growth than single protein source (Deshimaru and Shigueno, 1972; Conklin et al., 1977). According to New (1976) relatively higher amount of animal proteins than plant protein sources gave better results in the mixed diet. The improved performance of mixed diet is mainly because of the fact that neither animal protein source nor plant protein source can provide all the essential amino acids in adequate levels. While the animal protein sources are deficient in sulphur containing amino acids, the plant proteins are deficient in lysine. The deficiency may be overcome by mixing of both the sources of protein

(Shigueno, 1972). Balazs *et al.* (1974) reported better growth rate in *M. rosenbergii* with a mixture of soyabean meal and tuna meal compared to soyabean or tuna meal used individually. New (1976) also reported better growth performance in *M. rosenbergii* using mixture of two or more portein sources than when used individually. Zein-Eldin and Corliss (1976) reported that in the mixed diet, animal and plant source ingredients are to be included in correct proportions to obtain better growth and survival of penaeid prawn, since interactions of various dietary components affect quality or palatability of the feed. Nelson *et al.* (1977) showed that the assimilation rates of juvenile prawn *M. rosenbergii* fed on mixed diet were low compared to that based on animal source, but higher than prawns fed on plany based diet only. Boghen and Castell (1980) reported that mixture of shrimp meal and fish meal (marine protein sources) were found to cover the protein needs of juvenile lobsters. Chen *et al.* (1985) observed improved performance of feed containing a mixture of animal protein source and plant protein source in *P. setiferus* and *P. vannamei.* Gopal (1986) also reported higher growth rates in *P. indicus* juveniles fed on diets containing both animal and plant protein sources than when they were used individually. Pascual (1988) reported higher growth rate in *P. monodon* juveniles when 10% Ipil - ipil leaf (*Leucaena leucocephala*) meal was incorporated in a diet containing animal protein source. Similar result was also reported when 35 to 45% soyabean meal was incorporated in a diet containing only animal protein source (Pascual, 1988).

2.4.4 Single cell protein sources

Single cell protein sources include a wide range of algae, fungi (including yeast) and bacteria, which are produced by fermentation process, for use in an animal feed. By far, the greatest research efforts has been centred on the use of marine yeast SCP for feeding aquatic organisms. The discovery of marine yeast goes back to 1894, when Fisher separated red and white yeast from the Atlantic Ocean and identified them as *Torula* sp. and *Mycoderma* sp. Following Fisher's discovery, many other workers such as Hunter (1920), Bhat and Kachwalla, (1955); Suehiro, (1960) and Uden and Fell (1968) isolated marine yeast from different sources, Viz. sea water, marine deposits, seaweeds etc. In 1971, Kawano and Ohsawa started mass culture of marine strains of *Saccharomyces* sp. and tested its practical applications in mariculture. The utilization of marine yeast for feeding the larvae of *P. japonicus* has been demonstrated by Furukawa (1972), Furukawa and Hidaka (1973) and Furukawa *et al* (1973). Tiews *et al.* (1979) reported that the alkane yeast (*Candida lipolitica*) was found to produce equivalent growth to fish meal based ration in rainbow trout. But Atack and Matty (1979) found reduced weight gain and feed efficiency for alkane yeast compared to fish meal ration in rainbow trout. Nevertheless, in practice alkane yeast has been successfully incorporated in salmonid rations replacing upto 25-50% of the fish meal component with no loss in growth or feed efficiency (Tiews *et al.*, 1979; Beck *et al.*, 1979; Mahnken, *et al.*, 1980; Spinelli *et al.*, 1979). A combination of *Turulopsis utilis* and *Endomycopsis fubuliger* (Symba yeast) and the mould *Pekilomyces varioti* has also been successfully used to replace 50% of the fish meal protein in the diet of Atlantic Salmon (Bergstorm, 1979).

Similar results have also been obtained with bacterial SCP and in particular the bacterium *Methylophilus methylotrophus* indicating that methanol bacteria SCP can replace upto 75% of the fish meal protein in salmonid ration (Beck et al., 1979; Spinelli et al., 1979; Tacon et al., 1983) and upto 50% of the fish meal protein in a tilapia production diet (Viola and Arieli, 1984). Atack et al. (1979) reported that bacterial SCP was found to have higher nutritive value for common carp than alkane yeast SCP, when used as sole source of dietary proteins. Despite the common use of unicellular algae and yeast SCP as a live food organism for a variety of fish and crustacean larvae (Appelbaum, 1979; Watanabe et al., 1983) respectively, few studies have been reported on the direct use of dried algal meal in compounded feeds of fish and prawn (Stanley and Jones, 1976). In general, dried algae SCP has been found to have a lower food value for fish than either yeast SCP, bacterial SCP or fish meal (Atack and Matty, 1979). However, studies of Appler and Jauncey (1983) with Oreochromis nilotica and Hepher et al. (1979) with common carp indicate that certain dried algal meals may offer particular promise as potential dietary replacement for fish meal within practical food rations.

2.5 Oxygen consumption, Ammonia excretion and O : N ratios

Determination of metabolic rates by measuring oxygen consumption is of prime importance in quantifying the energy expenditure of an animal. Oxygen consumption of crustaceans has been studied by several workers (Subrahmanyam, 1957 & 1962; Rao, 1958; Kutty, 1967 and Kutty et al., 1971). Laxminarayana and Kutty (1982) reported reduced oxygen consumption under hopoxic conditions.

Ammonia excretion rates in prawns were also studied by many workers

(Wickens, 1976; Nelson et al., 1977; Stern and Cohen, 1982). Wickens (1976) reported an ammonia excretion rate of 0.25 to 0.85 mg N/g/day for juvenile M. rosenbergii. Seven fold increase in ammonia excretion rate was found in heavily fed prawn than starved ones (Nelson et al., 1977). Stern and Cohen (1982) reported an increased ammonia excretion during intermoult stages than premoult to moult stages. Gopal (1986) reported less ammonia excretion rate for P. indicus juveniles fed with animal protein sources than plant protein sources.

Oxygen to nitrogen ratios have been used as an index of the energy substrate utilization by many workers (Bayne and Scallad, 1977; Cupuzzo and Lancaster, 1979; Ragnault, 1981). Capuzzo and Lancaster (1981) reported decreased O : N ration in Homarus americanus which they attributed to increased protein catabolism. Regnault (1981) observed a reduction in O : N ratios in Crangon crangon when it is subjected to continued exposure to stress conditions. Seasonal variations in O : N ratios in P. varians have been indicated by Snow and Williams (1971), who reported lower values in winter and higher values in summer.

2.6 Ration size and feeding frequency

The ration size and the frequency with which animals consume feeds have been shown to have substantial effects on their metabolism (Cohen et al., 1963). It is a common practice in hatcheries to vary the feeding regime to improve the growth and survival rates of the larvae (Ishiwata, 1969; Kono and Nose, 1971).

The quantity of feed offered to an animal is generally based on the body weight and ranges from 5 to 100% dry diet of wet body weight depending on the size of the animal (Subrahmanyam and Oppenhemier, 1970; Venketaramiah et al., 1972). In nutritional studies it is important and essential to ensure that the food is always available for feeding ad libitum. However, feeding level should be regulated according to the consumption and the diet left over. Feeding animals in divided doses twice or thrice a day was found to contribute towards better growth and conversion efficiencies than feeding the entire dose once in a day (Primavera et al., 1979; Chua and Teng, 1982; Cuzon et al., 1982).

In intensive monoculture farming systems of shrimp and other crustaceans in controlled tanks, application of optimum ration and feeding frequency have much significance both for improving production and reducing the cost of production. If the feeding frequency is restricted to once daily, the left over feed disintegrates, microorganisms attack the sedimented, suspended and dissolved components which result in general fouling. To prevent these problems, the culturists increase the frequency of daily feeding to four or more meals and reduce the quantity per meal (Sedgwick, 1979). Frequent feeding has been reported to improve the growth of fish (Kono and Nose, 1971; Andrews and Page, 1975; Grayton and Beamish, 1977; Chua and Teng, 1978; Arunachalam and Reddy, 1981; Sampath, 1984) and prawns (Sedgwick, 1979; Marian et al., 1987; Sampath and Srithar, 1987). Marian et al. (1987) reported three meals a day as optimum for juvenile Macrobrachium lamerei. Sampath and Srithar (1987) report that in P. monodon rate of feeding, absorption and production increased as frequency of feeding was raised from once in five days to twice a day. However increasing the frequency of feeding beyond a level may not be economical.

According to Sick and Baptist (1973), factors such as age, and size of the animal, environmental parameters such as salinity, oxygen, temperature and pH of the water are known to influence the food consumption. They reported that consumption rates of P. setiferus were directly proportional to the animal size and length of exposure to any particular feed. Similar observations were also reported in P. duorarum (Sick et al., 1973).

2.7 Water stability of the artificial feeds

As the supplementary feed has to be provided in the water column to the aquatic animal, its water stability plays a major role in its performance. The problem of water stability for shrimp feed was first reported by Forster and Beard in 1969. Hastings et al. (1971) demonstrated that standard hard pellets lost 10% of their formulated feed stuff every 10 minutes after immersion in moving water. The physical instability of feed pellets and leaching out of specific hydrosoluble nutrients are serious problems in crustacean cultures since, most cultured species are demersal, continuous feeders and grasp feed pellets with pincer like appendages and mastigate externally (Forster, 1971; Zein-Eldin and Meyers, 1973; Pravasoli, 1976).

Water stability of crustacean diets using different binding materials have been studied by many workers (Meyers et al., 1972; Meyers and Zein-Eldin, 1972; Balazs et al., 1973; Meyers and Brand, 1975; Meyers, 1980). Prominent binding materials used in the prawn feeds are agar agar, alginates (sodium) gelatin, gums, carboxymethyl cellulose, polyvinyl alcohol, Carrageanan (polysaccharide), zein (corn protein) and starch (Kanazawa *et al.*, 1970; Forster, 1971; Grajcer and Neal, 1972; Meyers *et al.*, 1972; Sick *et al.*, 1972; Forster and Beard, 1973; Balazs *et al.*, 1973; Sick and Harris, 1974). Selection of binding agent has to be done carefully as some of the binders are very expensive and can considerably enhance the cost of the final feed. Natural feed ingredients such as wheat gluten, rice flour and tapioca are also used as binders besides being feed components. While tapioca which contains more than 70% starch is a good source of carbohydrate in the feed, it is also found to be a potential binding agent for prawn feed (Ahamad Ali, 1986). According to him, feed pellets prepared with 20% topioca were stable under water for more than 6 hours. Its binding quality compared well with the binding quality of agar agar, sodium alginate, and polyvinyl alcohol and was superior to that of pure starch.

Ingredient composition, nature of ingredient, type of processing and moisture content are known to influence feed stability (Hastings, 1971; Kainz, 1977). According to Stivers (1971), the degree of stability is dependent on the gelatinization of starch content of the feed during cooking. Hastings (1971), has stated that higher fat content affects gelatinization, thereby reducing the stability of the feed. Jayaram and Shetty (1981), are of opinion that the degree of stability depends on the gelatinisable material present in the feed. It was reported that grinding raw materials of the feed to about 200 microns has resulted in increased water stability of the pellets, enhanced digestability and best food conversion ratio in the prawn *P. indicus* (Rani, 1984).

Better stability of the prawn feeds is always desirable to improve their utilization, reducing the quantity of feed to be supplied to the prawn as well as reducing the cost of operation. But too much stability of the pellets is also undesirable, since the nutrients may get tied up so tightly that they become unavailable to the organism in addition to the higher cost of such feed (Balazs et al., 1973).

2.8 Keeping quality of artificial diets

The shelf life of artificial shrimp feeds are dependent on the type of processing, storage, temperature and moisture content of the diet (Hastings, 1971; Hilton et al., 1977). During prolonged and improper storage, adverse physical conditions (moisture, heat, light) and microorganisms (mould, bacteria, yeast) can cause deterioration of feed quality, resulting in decreased palatability and nutritive value including deterioration of amino acid (Chow, 1980).

A good quality feed is one which is able to maintain its nutritional status for a few weeks of storage. Fowler and Banks (1967) have found no alterations in the nutritional status of the pellets stored at room temperature for a few weeks, but storage for greater periods had deleterious effect on the growth performance of fish. Jayaram and Shetty (1980) reported increase in moisture content of feed after 3 months storage period and attributed it to the presence of hygroscopic ingredients in the feeds, and also difference in the porosity of feed. The percentage decrease in crude protein and crude fat during storage period have been reported by Jayaram and Shetty (1980) and Venugopal and Keshavanath (1984). The reason for this decrease in crude protein is due to an increase in moisture level which would have facilitated the breakdown of proteins as well as the degree of unsaturated fatty acids in the feed (Jayaram and Shetty, 1980). The rate of oxidation of fat is greatly influenced by the storage temperature and rise of 10°C approximately doubles the rate (Kulikov, 1978). Oxidation of fat gives rise to a feed stuff of lower biological value (Rumsey, 1980), that may cause a reduction in growth of the animal fed on that diet (Stuart et al., 1985). The use of deteriorated feeds also can result in increased disease outbreaks such as lipoid liver degeneration (Roberts, 1978). The percentage decrease on the water stability of the feeds stored for 3 months have also been reported by Venugopal and Keshavananth (1984). Cho et al. (1985) reported that during storage feed quality diminishes due to waste, chemical deterioration and infestation by microbes or insects. Proper storage of feeds in air tight containers can reduce the problems to some extend.

Oxidation can be reduced by lowering the storage temperature and controlling storage conditions (Kulikov, 1978). Hardy (1980) reported that rate of oxidation of the feed can also be reduced by reducing the temperature and oxygen levels during storage. Natural antioxidants such as tocopherol, chemical antioxidants like butylated hydroxy anizole (BHA), and butylated hydroxy toluene (BHT) can be used to prevent lipid oxidation in feeds. Fungal growth in feed can be prevented by the use of potassium metabisulphite, sorbic acid, benzoic acid, and propionic acid.

Dela Cruz, et al. (1989) recomended that feeds should not be stored for more than fifteen days during summer months, when the temperature ranges from 28 - 31°C. Feeds can be stored for upto 30 - 35 days during cooler months (10 - 20°C). They also suggested that a periodic check of peroxide levels should be done inorder to detect early stage of feed deterioration.

III. MATERIALS AND METHODS

III MATERIALS AND METHODS

Experiments have been conducted to evaluate the efficiency and keeping quality of diets based on different protein sources (animal, plant and single cell protein) in the juveniles of P. monodon. The optimum food ration and feeding frequency of the selected diet, which gave best growth performance in the first experiment were also studied.

3.1 Preparation of the Feed

3.1.1 Feed ingredients

Animal, plant and single cell protein sources used for the experiment were as follows:

Animal origin : clam meal, prawn meal and slaughter house waste meal*

Plant origin : soyabean meal

Single cell origin : marine yeast

Other ingredients used in the pelleted feeds were ground nut oil cake, rice bran, tapioca powder, sardine oil and mineral mix.

Prawn meal, clam meal, and slaughter house waste meal were prepared by sundrying and powdering them. The prawn meal was prepared from metapenaeid prawn (M. dobsoni) obtained from the farm of the College of Fisheries, whereas the clam meat and slaughter house waste were obtained from the local market. Marine yeast was obtained from the Microbiology lab

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* Prepared from Beef intestine, stomach, liver, discarded meat etc.

of the Processing Department, College of Fisheries and sardine oil obtained from Fishery Byproduct Section, CIFT.* Minerals were supplemented through salt mixture USP IX (SISCO RESEARCH LABORATORIES PVT LTD, BOMBAY). All the ingredients were finely powdered and passed through a 500 µm mesh and stored in polythene bags.

3.1.2 Proximate composition of Feed ingredients

Proximate composition of all the feed ingredients was analysed prior to feed formulation.

The moisture level was estimated by heating the sample to 105°C for 30 minutes and then drying at 65°C till a constant weight was obtained, following the method of Boyd (1979). The crude protein content was estimated by Microkjeldahl's method (AOAC, 1975). The nitrogen content was multiplied by the factor 6.25, to arrive at crude protein content. Crude fat was extracted using petroleum ether (B.P. 40°C - 60°C) in a Soxhlet extraction apparatus for 16 hrs. The crude fibre content was estimated by the method of Pearson (1976), while ash content was determined by burning the sample at 550°C ± 10°C for 6 hrs in a muffle furnace. The carbohydrate content (nitrogen free extract, NFE) was found out by Hastings (1976) difference method.

NFE = 100 - (% crude protein on dry weight basis + % crude fat on dry weight basis + % crude fibre on dry weight basis + % ash).

3.1.3 Formulation and Processing of test diets

Five types of pelleted feeds were formulated maintaining their protein

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* Central Institute of Fisheries Technology.

level at 40%. They were:

1. CM (clam meal as the major protein source)
2. PM (prawn meal as the major protein source)
3. SW (Slaughter house waste meal as the major protein source)
4. SM (soyabean meal as the major protein source) and
5. MY (marine yeast, as the major protein source).

Table 1 gives the proportion of ingredients used for the preparation of different pelleted feeds and nutrient composition of the diets. Each type of pelleted feed was prepared separately by mixing the required quantity of ingredients. The mixture was hand kneaded with sufficient water (1 : 1.25 W/V) to get a soft dough. The dough was cooked for 30 minutes in an autoclave at ambient pressure. The cooked dough was cooled, salt mixture and sardine oil were then added, mixed, pelletized and dried at 60°C overnight in a hot air oven to a moisture level $\leq$ 10%. The pellets were packed separately in plastic bags and stored free from moisture and sunlight.

3.1.4 Proximate composition of formulated feeds

The proximate analysis of the formulated feeds were carried out immediately after their preparation and again at the end of 4 month's storage period. Methodology employed was the same as that of ingredients. Calcium and phosphorus content of the pelleted feeds were analysed by the method of AOAC (1970).

Table 1. Proportion of different ingredients used in the preparation of pelleted feeds.

Ingredients	CM	PM	SW	SM	MY
Clam meal	62	-	-	-	-
Prawn meal	-	65	-	-	-
Slaughter house waste meal	-	-	59	-	-
Soyabean meal	-	-	-	67	-
Marine yeast	-	-	-	-	72
Groundnut oil cake	15	15	15	15	15
Rice bran	10	10	10	10	5
Tapioca Powder	11	8	14	6	6
Sardine oil	1	1	1	1	1
Mineral mix*	1	1	1	1	1
Total	100	100	100	100	100

* Mineral mix (Salt mixture USP IX - SISCO Research Lab. Pvt. Ltd. Bombay)

The energy value of each feed was found out by multiplying by factors 9.46 for fat, 4.18 for carbohydrate, and 4.32 for proteins (Bages and Sloane, 1981). The energy values were denoted as K. Calories/g.

3.2 Sinking rate of pelleted feeds

The sinking rate of all pelleted feeds in dry conditions was determined in an aquarium tank measuring 1.25 x 0.5 x 0.5 m, soon after feed preparation and also after 4 months storage period. Ten uniform sized pellets of about 1 cm were gently dropped into the aquarium and the time taken by them to traverse the water column was recorded using a stop watch. The average time taken by each type of pellet was found out separately and average sinking rate of feeds were expressed in cm/sec.

3.3 Water stability of pelleted feeds

Water stability (% dry matter obtained after exposing the pellets in water for 3 hrs) was determined by the method of Jayaram and Shetty (1981).

3.4 Total heterotrophic bacteria and fungi

Total viable bacterial count was determined following standard pour plate method using the plate count agar (Tryptone 0.5%, Yeast extract 0.25%, Dextrose 0.1%, Agar 1.5%, pH 7 ± 0.2). Total viable fungal count was taken in the same way using sabaraud dextrose agar (Mycological Peptone - 1%, Dextrose - 4%, Agar - 1.5%, pH - 5.6 ± 0.2).

3.5 Keeping quality of pelleted feeds

Before and after storage (4 months), samples from each type of feed were analysed for proximate composition, water stability, sinking rate, total bacterial count and total fungal count.

3.6 Experimental animals

Post larvae of Penaeus monodon belonging to the same brood stock were obtained from the Regional Shrimp hatchery at Azhicode and transported in polythene bags of 10 litre capacity half filled with fresh filtered sea water of 25 ppt salinity under oxygen packing. The postlarvae were then introduced into cement cisterns of 350 litre capacity at the rate of 100 to 150 animals per tank. The water salinity in these cisterns were maintained at 20 ppt. The postlarvae were fed with compounded pellet diets to obtain desired early juveniles (Average weight about 200 mg) for experimentation.

3.7 Experimental tanks and water management

Experiments were carried out in 50 litre plastic tanks (60 x 40 x 40 cm), in which aeration was provided with the help of an air compressor using air stones.

Brackish water with salinity of 25 ppt collected from the backwater near College campus, was filtered twice, using bolting silk and filled in cement cisterns. The salinity of the water was then adjusted to 20‰ by diluting with fresh water and was used for rearing animals in feeding experiments. One third of the water was replaced daily by water of the same salinity, while complete water was replaced in the tanks once in a week.

3.8 Study to evaluate the protein source

Juveniles prawns of mean total length (3.23 ± 0.25 cm) and weight (0.195 ± 0.02 gms) were used for the experimental work. The prawns were allowed to starve for 24 hours prior to the experiment. The total length of apparently healthy animals were measured to the nearest centimetre from tip of rostrum to the tip of telson. The animals were then blotted dry carefully between the folds of filter paper weighed in an electronic balance to the nearest milligram and were transferred to tanks. Before starting the experiment about 15 prawns were measured, weighed and left for drying in an oven at 40°C for 48 hrs. Dried prawns were then weighed and the body protein content of these were analysed (AOAC, 1975).

Fifteen numbers of P. monodon juveniles were introduced in each tank. For each treatment of feed four replicates were maintained. Hence 60 animals were used for each treatment. The duration of the experiment was for a period of 42 days.

The juvenile prawns were fed with the experimental diets ad libitum twice daily in petri dishes kept at the bottom of the tanks as suggested by Subramanyam and Oppenheimer (1970). Feeding was done in the morning and evening. Before feeding, left over feed was recovered and adhering salt was washed gently and dried in an oven at 60°C to constant weight.

3.8.1 Determination of digestability coefficient

The animals were allowed to fully acclimatize to the diet, and no faeces were analysed during the first week, after that, faeces were collected for a period of 20 days by slow siphoning of the water through a narrow polythene tube and collected on a bolting silk. The faecal matter were washed in distilled water to remove adhering salt, then transferred to pre-weighed beaker and weighed after drying in an oven at 60°C to constant weight. The dry faeces were homogenized in a mortar and used for protein estimation (AOAC, 1975).

3.8.2 Monitoring of Physico-Chemical parameters of water

Water temperature was recorded twice daily, morning at 8 A.M. and evening at 6 P.M., using graduated mercury thermometer with an accuracy of 0.1°C. The dissolved oxygen content in water samples was determined daily employing Winkler's method (Strickland and Parsons, 1972). Salinity of water in the experimental tanks was determined thrice a week using refractometer. pH of the water from the experimental tanks was determined thrice a week using digital pH meter with an accuracy of 0.01 (ELICO - Model L1-122).

3.8.3 Recording of the data

3.8.3.1 Survival rate

The percentage survival was determined as follows:

$$\text{Percentage survival} = \frac{\text{Initial number - Final number}}{\text{Initial number}} \times 100$$

3.8.3.2 Growth

Growth of the animal in length and weight was calculated by the following formula.

$$\text{Growth \%} = \frac{\text{Final measurement - Initial measurement}}{\text{Initial measurement}} \times 100$$

3.8.3.3 Specific growth rate

$$\text{SGR} = \frac{\log_e w_2 - \log_e w_1}{t_2 - t_1} \times 100$$

Where w_1 = weight at time t_1

w_2 = weight at time t_2

The calculated value gives the average percentage increase in body weight per day over 42 days.

3.8.3.4 Food conversion ratio (FCR)

FCR of the diet was calculated using increase in live weight and food consumed by the following formula.

$$\text{ECR} = \frac{\text{Av. wt. of food consumed in dry weight}}{\text{Av. live weight gain}}$$

3.8.3.5 Protein efficiency ratio (PER)

PER was calculated by the formula

$$\text{PER} = \frac{\text{Av. live weight gain}}{\text{Av. protein consumed}}$$

3.8.3.6 Protein digestability coefficient

$$= \frac{\text{Protein digested}}{\text{Protein ingested}} \times 100 \qquad \frac{[(F \times P) - (E \times Q)]}{(F \times P)}$$

F = Food consumed, E = Excreta produced, P = Protein in feed,
Q = Protein in excreta

3.8.4 Biochemical composition of carcass

On termination of the experiment, the carcass of the prawn was analysed for moisture, crude protein crude fat, ash, carbohydrate, calcium and phosphorus, following the standard procedures mentioned earlier, inorder to evaluate the best protein source.

3.8.5 Estimation of oxygen : nitrogen ratios

Intermoult prawns fed for 42 days, on different diets were selected in triplicates from each of the treatments for experimental study. The prawns were acclimated in the respirometer chamber for one hour before readings were taken and the chamber was flushed with the respective growth media throughout the period of equilibration. At the beginning of each experiment water sample was drawn for estimation of dissolved oxygen and then the respirometer was closed. All the respirometers were placed in a water bath and maintained at a temperature 30 ± 1°C. After one hour, water sample was drawn for oxygen estimation. The difference was considered to be the oxygen consumed by animal during the period of experiment. After completion of the experiment, the prawns were blotted dry with filter paper and weighed. The results are expressed in mg O_2/hr and the metabolic rates (mg O_2/g/hr) were also calculated.

Ammonia excretion was determined using water samples drawn from the respirometer chamber prior to and after respirometry. The results are expressed as mg NH_3 - N/g/hr. All the readings were taken during the dark phase of photoperiod in order to assure uniformity of experimental conditions between each runs.

Using oxygen consumption and ammonia excretion rates O : N ratios of individual prawn fed with different protein sources were estimated following the method given by Bayne et al., 1985.

3.9 Study to determine the optimum food ration

Optimum food ration for the selected diet, which gave the best growth performance and conversion efficiency in the first experiment was determined. Fifteen numbers of the juveniles of P. monodon (Av. length 3.3 ± 0.01 cms, Av. wt. 0.209 ± 0.06 g) were introduced in each experimental tank. Food rations with 0, 3, 6, 9, 12, 15, 18 and 21 percentages of the body weight were given to the prawns. Each treatment had 3 replications. The experiment was conducted for a period of 21 days and specific growth rate, survival (%) and food consumption rate were determined.

3.10 Study to determine optimum feeding frequency

Optimum feeding frequency of the selected diet was determined as the third part of the study. Here also 15 numbers of P. monodon juveniles (Av. length 3.2 ± 0.02 cms, Av. wt. 0.214 ± 0.02 gms) were introduced in each tank. The prawns were fed on the selected diet at the rate of 15% of the body weight which was the maximum ration obtained in the second experiment at different frequencies viz.

Once a day

a) day alone (0800 hrs)
b) Night alone (2000 hrs)

Twice a day

c) day and night (0800 hrs and 2000 hrs)
d) day and noon (0800 hrs and 1400 hrs)
e) Noon and night (1400 hrs and 2000 hrs)

Thrice a day

f) Morning, noon and night (0800 hrs, 1400 hrs & 2000 hrs)

Four times a day

g) 0800 hrs, 1100 hrs, 1400 hrs, 1700 hrs & 2000 hrs.

The experiment was conducted for a period of 28 days and observations on specific growth rate, food consumption and survival were determined.

3.11 Statistical Analysis

Analysis of variance (Snedecor and Cochran, 1968) was carried out for the collected data. Ratios and percentage values (x) were transformed into Arc sin values ($\text{Sin}^{-1}\sqrt{\frac{x}{100}}$) for analysis.

IV. RESULTS

IV RESULTS

4.1 Proximate Composition of Feed Ingredients and Formulated Feeds

4.1.1 Feed Ingredients

The proximate composition of the feed ingredients used in the formulation of the experimental diets is shown in Table 2.

The moisture content of the ingredients viz, clam meal, prawn meal, slaughter house waste meal, soybean meal, marine yeast, ground nut oil cake, ranged from 4.04 - 13.68%, the maximum in marine yeast (13.68%) and minimum in soybean (4.04%).

The highest crude protein content was recorded in slaughter house waste meal (56.36%) while it was lowest in the case of tapioca powder (7.1%). The protein percentage of other ingredients viz. clam meal, prawn meal, soybean meal, marine yeast, ground nut oil cake and rice bran were 53.5%, 50.8%, 48.8%, 46.9%, 32.9% and 13.9% respectively.

The crude fat content of the ingredients ranged from 0.75% (rice bran) to 12.6% (slaughter house waste meal). Tapioca powder contained the highest nitrogen free extract (76.55%), whereas prawn meal had the least amount (1.2%). The highest value of crude fibre (21.42%) was recorded in rice bran, while the lowest value (0.51%) was observed in clam meal. The percentage of ash varied from 1.45% to 36.15%, the highest being in rice bran and lowest in tapioca flour.

Table 2. Proximate composition of the ingredients used in feed formulation

Ingredients	Moisture (%)	Crude Protein (%)	Crude fat (%)	Crude fibre (%)	Carbohydrate (N - free extract) (%)	Ash (%)
Clam meal	8.79	53.5	10.8	0.51	16.8	9.6
Prawn meal	11.50	50.8	9.7	3.2	5.6	19.2
Slaughter house waste meal	12.06	56.36	12.6	0.8	1.2	16.98
Soyabean meal	4.04	48.8	4.32	4.06	30.22	8.56
Marine yeast	13.68	46.9	2.4	0.6	25.23	11.19
Groundnut oil cake	10.12	32.9	7.56	6.12	36.27	7.03
Rice bran	7.32	13.9	0.75	21.42	20.46	36.15
Tapioca	10.04	7.1	0.80	4.06	76.55	1.45

4.1.2 Formulated Pelleted Feeds

4.1.2.1 Initial Analysis

Data on the proximate composition of all the formulated feeds are presented in Table 3. The moisture content of the five feeds ranged between 7.86 and 9.82% while the crude protein content was between 39.76% (MY) and 40.30% (SW). The percentage of nitrogen free extract was the highest in the pellet SM (29.38%) and lowest in the pellet PM (19.80%). The crude fibre content ranged from 3.76% (MY) to 7.02% (SM) and ash content between 10.42% (SM) and 15.43% (PM). The calcium content of the pelleted feeds ranged between 298.06 mg % (SM) and 392.26 mg % (MY), while the range of phosphorus was found to be 530.16 mg % (SM) and 844.20 mg % (MY). The caloric values of the five test diets were almost similar, ranging from 3.34 to 3.72 K.cal/g. Hence the test diets were more or less isocaloric and isonitrogenous.

4.1.2.2 Analysis after Storage

The data of the experiment to study the effect of storage on the keeping quality of pelleted feeds after 4 months of storage are presented in Table 4.

The moisture content of the test diets ranged from 9.06 to 13.32%, the highest being in MY while the lowest in SM respectively. The crude protein content ranged between 35.56 (MY) to 37.57% (SM). The highest value of crude fat was observed in pellet SW (8.71%) and the lowest in the pellet MY (3.90%). The values of crude fibre content were 4.12, 5.86, 4.07, 6.16 and 3.36% for pellets CM, PM, SW, SM and MY respectively.

Table 3. Proximate composition of formulated feeds

Parameters*	Diets				
	CM	PM	SW	SM	MY
Moisture (%)	8.32	9.74	9.20	7.86	9.82
Crude protein (%)	40.24	40.01	40.30	40.17	39.76
Crude fat (%)	8.99	8.56	9.72	5.15	4.76
Crude fibre (%)	4.82	6.46	4.89	7.02	3.76
Cabohydrate (%) (N - free extract)	27.07	19.80	21.57	29.38	28.09
Ash (%)	10.56	15.43	14.32	10.42	13.61
Calcium (mg %)	324.60	386.07	346.67	298.06	392.26
Phosphorus (mg %)	616.93	826.42	779.93	530.16	844.20
Caloric value (K. calories/g)	3.72	3.37	3.56	3.45	3.34

* Average of four values

The ash content was maximum in pellets PM (14.08%) while it was minimum in pellet SM (9.56%). The nitrogen free extract ranged from 23.82% to 32.7%, the highest and the lowest values being in pellets PM and SM, respectively.

4.2 Sinking Rate of Pellets

Pellet SM exhibited the highest sinking rate (4.62 cm/sec) while lower rates were observed in the case of PM and MY (Table 4). The average sinking rate of pellet CM, was 4.82 cm/sec and that of SW was 5.71 cm/sec.

Following storage, an increase in average sinking rate was observed in all feeds. The highest values was recorded in the case of MY (7.96 cm/sec). It was followed by pellet PM and SW respectively, the values being 6.82 cm/sec and 6.41 cm/sec. The lowest value was recorded in the case of pellet SM (5.02 cm/sec).

4.3 Water Stability

The results of the stability test for different pellets are shown in Table 4. The most stable feed at the end of three hours was pellet SM (91.13%), followed by CM (87.26%) and SW (83.02%). The lowest stability was observed for the pellet MY (78.94%).

A decrease in average stability in all the feeds was seen after four months of storage (Table 4). The decrease in stability was maximum (-4.28%) in the case of pellet MY, and minimum (-2.22%) in pellet SM.

Table 4. Effect of storage on the keeping quality of pelleted feeds

Parameters*	Fresh	After 4 months storage	Percent difference	Fresh	After 4 months storage	Percent difference	Fresh	After 4 months storage	Percent difference	Fresh	After 4 months storage	Percent difference	Fresh	After 4 months storage	Percent difference
Average sinking rate cm/sec	4.82	5.44	-	5.85	6.82	-	5.71	6.41	-	4.62	5.02	-	6.94	7.86	-
Stability (%)	87.26	85.06	2.52	80.26	77.66	3.24	83.02	80.27	3.31	91.23	89.11	2.22	78.94	75.56	4.28
Moisture (%)	8.32	10.97	31.85	9.74	11.39	16.94	9.20	12.20	32.61	7.86	9.06	15.27	9.82	13.32	35.64
Crude protein (%)	40.24	36.99	8.08	40.01	37.05	7.40	40.30	36.68	8.98	40.17	37.57	6.47	39.76	35.56	10.56
Crud fat (%)	8.99	8.11	9.79	8.56	7.80	8.88	9.72	8.71	10.39	5.15	4.55	11.65	4.76	3.90	18.07
Ash (%)	10.56	9.76	7.58	15.43	14.08	8.75	14.32	13.82	3.49	10.42	9.56	4.41	13.81	13.21	4.34
Crude fibre (%)	4.82	4.12	14.52	6.46	5.86	13.45	4.89	4.07	16.77	7.02	6.16	12.25	3.76	3.36	10.64
Carbohydrate (%)	27.07	30.09	11.16	19.80	23.82	20.30	21.57	24.52	13.68	29.38	32.70	11.30	28.09	30.65	9.11
Fungal count (c.f.u/g)	0	5.2×10^7	-	0	2.99×10^5	-	0	3.20×10^8	-	0	1.09×10^5	-	0	2.91×10^9	-
Bacterial count (c.f.u./g)	1.51×10^5	8.26×10^6		4.05×10^5	6.20×10^6		7.8×10^5	9.2×10^6		2.90×10^5	3.15×10^6		2.5×10^6	5.10×10^7	

* Average of four values

4.4. Total Heterotrophic Bacteria and Fungi

Viable bacterial population (c.f.u/g) in the fresh pelleted feeds ranged between 1.51 x 10^5 and 2.5 x 10^6, the highest being recorded in pellet MY and the lowest in pellet CM respectively. No fungi could be recorded in fresh samples.

An increase in bacterial count was observed in different feeds after four months of storage. Pellet MY was found to harbour the highest bacterial population (5.1 x 10^7 c.f.u/g) and pellet CM the lowest (3.15 x 10^6 c.f.u/g). Subsequent to storage fungal populations ranging from 1.09 x 10^5 c.f.u/g to 2.91 x 10^9 c.f.u/g were recorded in pellets SM and MY respectively.

4.5 Evaluation of Relative Efficiencies of Natural protein sources

The details of various physicochemical parameters of the experimental tanks are presented below.

4.5.1 Physico-chemical Parameters

4.5.1.1 Temperature

Temperature in the experimental tanks ranged between 24.2°C to 29.8°C and average values recorded during each week are presented in Table 5.

4.5.1.2 pH

The pH of the water was found to be alkaline throughout the experiment in all the experimental tanks, its range being 7.62 to 8.40 (Table 6, Fig. 1).

Table 5. Temperature range (in °C) of the experimental tanks during the study of protein source evaluation

Temperature (°C)	WEEKS					
	1	2	3	4	5	6
Mean ± SD	27.37±1.49	27.13±1.13	28.16±1.06	27.32±1.45	28.07±1.52	27.46±1.16
Range	(26.0-29.5)	(26.5-29.2)	(26.7-28.8)	(25.2-29.8)	(25.4-28.9)	(24.2-29.8)

Table 6. Fluctuations of pH in the experimental tanks during the study of protein source evaluation (Range shown in parenthesis)

Treatments	WEEKS (mean ± SD)						
	0	1	2	3	4	5	6
CM	8.12 ± 0.22 (7.96 - 8.38)	8.02 ± 0.53 (7.86 - 8.42)	8.10 ± 0.23 (7.92 - 8.36)	8.06 ± 0.24 (7.80 - 8.28)	8.08 ± 0.16 (7.90 - 8.22)	8.20 ± 0.18 (8.10 - 8.42)	8.24 ± 0.14 (8.12 - 8.40)
PM	8.10 ± 0.16 (7.86 - 8.40)	8.01 ± 0.26 (7.72 - 8.22)	7.99 ± 0.25 (7.80 - 8.28)	8.17 ± 0.14 (8.01 - 8.36)	8.20 ± 0.19 (8.02 - 8.40)	8.16 ± 0.26 (8.12 - 8.28)	8.18 ± 0.16 (8.10 - 8.36)
SW	8.02 ± 0.18 (7.90 - 8.18)	7.96 ± 0.18 (7.68 - 8.12)	7.90 ± 0.26 (7.72 - 8.20)	7.82 ± 0.18 (7.70 - 8.16)	8.01 ± 0.14 (7.92 - 8.20)	8.06 ± 0.18 (7.98 - 8.18)	8.12 ± 0.16 (8.01 - 8.20)
SM	8.14 ± 0.26 (8.02 - 8.24)	8.12 ± 0.23 (8.02 - 8.36)	8.16 ± 0.16 (8.08 - 8.24)	8.24 ± 0.18 (8.16 - 8.28)	8.12 ± 0.18 (8.04 - 8.16)	8.20 ± 0.12 (8.12 - 8.26)	8.26 ± 0.16 (8.16 - 8.32)
MY	7.90 ± 0.18 (7.80 - 8.12)	7.76 ± 0.21 (7.62 - 8.02)	7.92 ± 0.16 (7.82 - 8.16)	7.90 ± 0.26 (7.86 - 8.14)	7.96 ± 0.14 (7.74 - 8.20)	8.16 ± 0.36 (8.01 - 8.26)	8.14 ± 0.16 (8.10 - 8.26)

Plate II

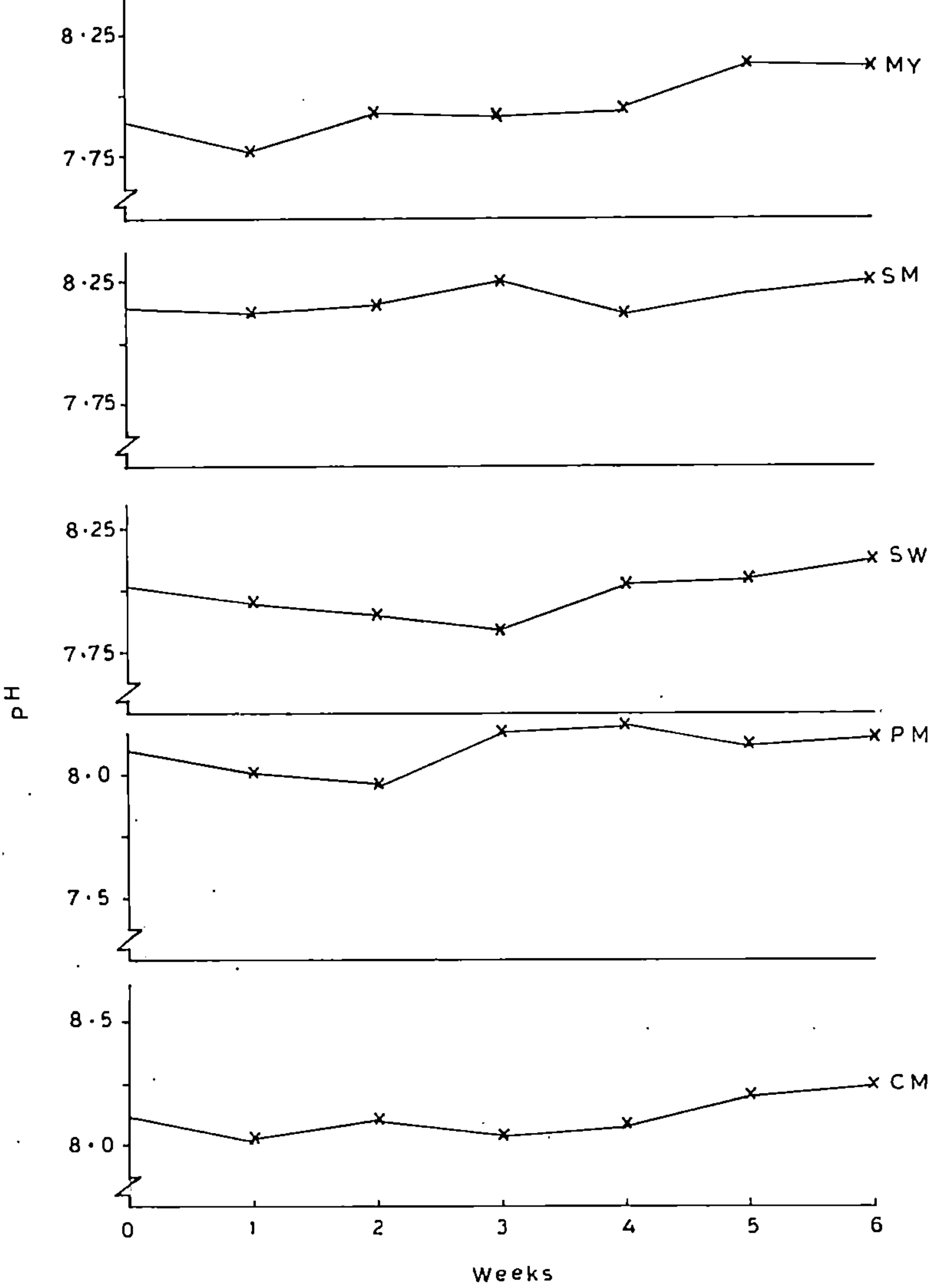

Fig. 1.

Table 7. Variations in dissolved oxygen content (ppm) of the experimental tanks during the study of protein source evaluation (Range shown in parenthesis)

Treatments	WEEKS (mean ± SD)						
	0	1	2	3	4	5	6
CM	6.64 ± 0.28 (6.26 - 6.83)	6.80 ± 0.28 (6.20 - 7.01)	6.26 ± 0.46 (6.02 - 6.60)	6.40 ± 0.34 (6.20 - 6.64)	6.56 ± 0.27 (6.18 - 6.90)	6.62 ± 0.26 (6.24 - 6.82)	6.40 ± 0.32 (6.16 - 6.68)
PM	5.76 ± 0.82 (4.44 - 7.22)	5.60 ± 1.20 (4.40 - 6.80)	5.73 ± 1.40 (4.40 - 7.20)	5.86 ± 1.02 (5.01 - 7.30)	5.68 ± 0.86 (4.20 - 7.01)	5.89 ± 1.02 (4.60 - 7.60)	5.70 ± 1.36 (4.5 - 7.10)
SW	7.43 ± 0.64 (7.20 - 7.64)	7.73 ± 0.50 (7.30 - 7.96)	7.56 ± 0.46 (7.01 - 8.01)	7.67 ± 0.42 (7.20 - 8.00)	7.67 ± 0.43 (7.20 - 8.01)	7.47 ± 0.64 (7.00 - 8.20)	7.36 ± 0.46 (7.10 - 7.60)
SM	6.01 ± 0.26 (5.20 - 6.48)	5.80 ± 0.80 (5.07 - 6.20)	5.96 ± 0.80 (5.50 - 6.26)	5.72 ± 0.25 (5.26 - 6.50)	6.02 ± 0.25 (5.26 - 6.45)	6.12 ± 0.46 (5.36 - 6.40)	6.06 ± 0.28 (5.32 - 6.50)
MY	7.12 ± 0.56 (6.72 - 7.46)	7.33 ± 0.80 (6.40 - 7.80)	7.27 ± 0.50 (.680 - 7.80)	7.67 ± 0.12 (7.60 - 7.80)	7.40 ± 0.20 (7.20 - 7.60)	7.27 ± 0.50 (6.80 - 7.80)	7.14 ± 0.46 (6.90 - 7.36)

Plate II

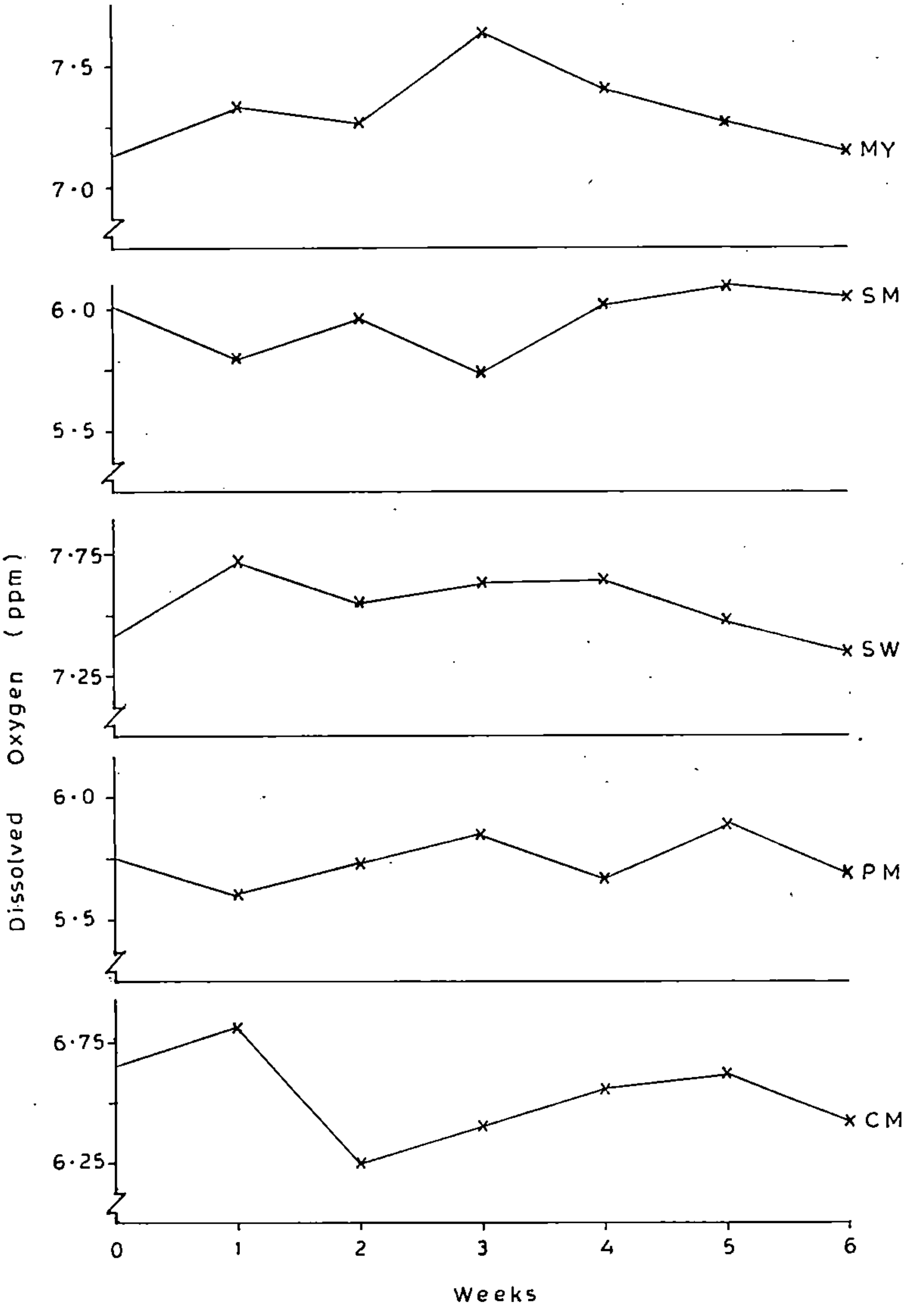

Fig. 2.

4.5.1.3 Dissolved Oxygen

The dissolved oxygen content of water recorded during the experimental period is shown in Table 7, Fig. 2. The average values in different treatments varied from 5.60 to 8.20 ppm.

4.5.2 Growth in relation to protein source

During the 42 days of experimental study, with five different feeds under four replication, the following observations were made.

4.5.2.1 Survival Percentage

The survival rates (%) of prawns recorded from various treatments are shown in Fig. 3. Analysis of variance of the data showed that the dietary protein sources significantly influence ($P < 0.01$) the survival of the prawns (ANOVA Table 1). Among the diets tested the highest survival (90%) was recorded in the prawn fed on diet containing clam meal followed by those fed on slaughter house waste meal (89%). The lowest % of survival was seen in prawns fed on diet containing soyabean meal (86%). The survival rates obtained for those fed on diet based on prawn meal are not significantly different ($P > 0.01$) from those based on marine yeast.

4.5.2.2 Growth

The net gain in length and wet weight of prawns and also the percentage growth are shown in Table 8, Fig. 5. Analysis of variance of the data show that the dietary protein sources significantly influence ($P < 0.01$) the above growth parameters (ANOVA Table 2). The prawns fed on diet containing

Plate III

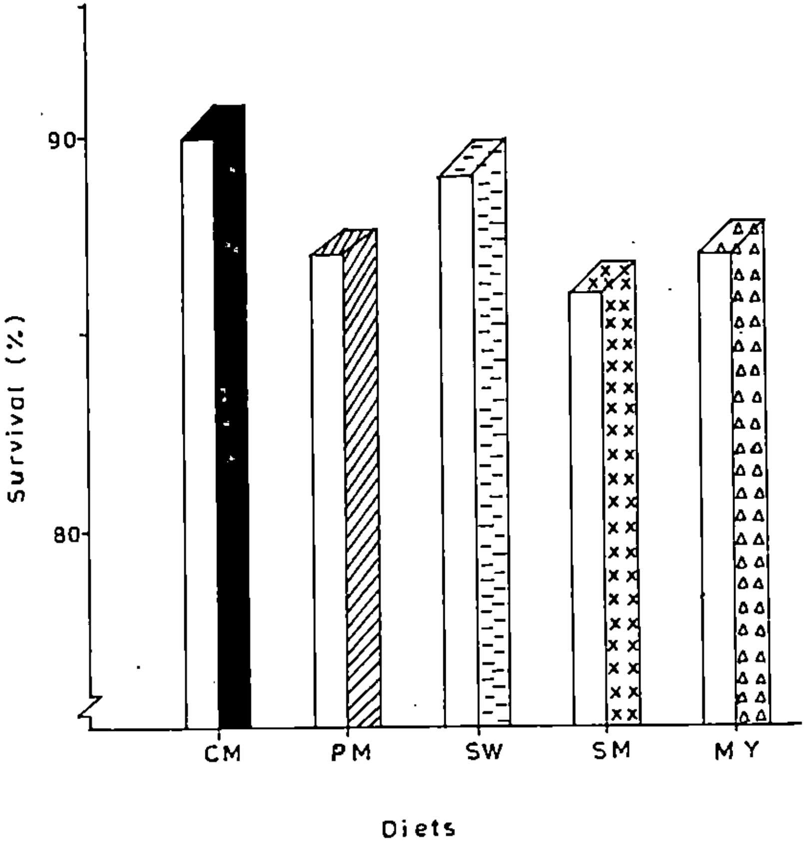

Fig. 3

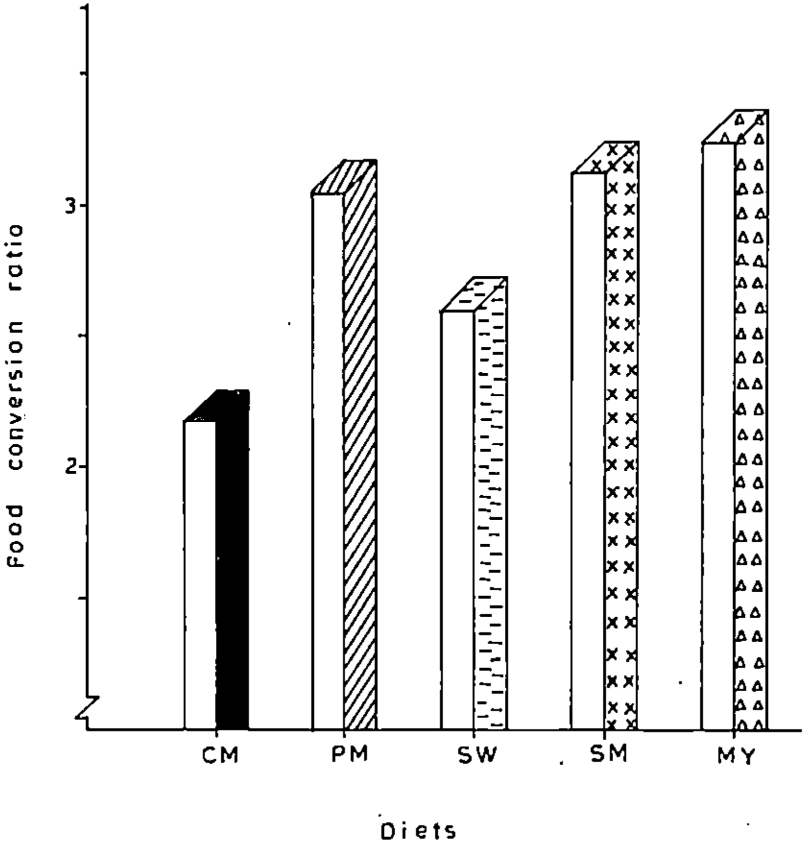

Fig. 4

ANOVA TABLE 1

Analysis of variance of the data on survival rate of *P. monodon* juveniles fed on diets based on different protein sources.

Source of variation	Degrees of freedom	Sum of squares	Mean sum of squares	F Value
Between diets	4	95.46	23.86	13.81**
Error	15	25.92	1.728	
Total	19	121.38		

Standard error of transformed treatment means = 0.929

Critical difference = 2.739

Comparison of transformed means based on critical difference

Diets	CM	SW	PM	MY	SM
Transformed mean values	71.57	70.63	68.86	68.86	68.03

Underscored means are not significantly different

** Significant at 1% level.

clam meal (CM) recorded the highest net gain in length (3.89 cm) among the various protein sources tested, while the lowest gain in length was observed in diet containing soybean meal (2.275). Gain in lengths recorded for prawns fed on diet based on slaughter house waste (SW) and marine yeast (MY) was 3.45 cm and 3.18 cm respectively. The net gain in weight of prawns fed on diets with plant protein source (SM) was found to be significantly less than ($P < 0.01$) those fed on diets based on animal protein sources (CM, PM and SW) or single cell protein source (MY) [ANOVA Table 2]. Among the feeds tested, diet based on clam meal (CM) gave the highest net gain in weight (2.035 gms) followed by diet based on slaughter house waste (1.475 gms). The net gain in weight recorded in prawns fed with prawn meal (PM) and slaughter house waste (SW) was not significantly different from each other.

4.5.2.3 Specific Growth Rate

Significant influence ($P < 0.01$) of dietary protein sources was observed on the specific growth rate of prawns (Table 9, Fig. 6, ANOVA Table 3). The prawns fed on clam meal based diet recorded the highest SGR (5.78%) followed by diet based on slaughter house waste (4.99%) and the lowest rate (3.63%) was recorded in those fed on diet containing soybean meal. S.G.R. values recorded in prawns fed on diets based on marine yeast and prawn meal were 4.53 and 4.34% respectively.

4.5.2.4 Food Conversion Ratio

The food conversion ratio (FCR) recorded in the study, (Table 10, Fig. 4) showed significant variation ($P < 0.01$) between diets from various

Table 8. Average weight (g) and length (cm) attained by P. monodon juveniles fed on diets based on different protein sources for a period of 42 days (values in the parenthesis is the average of four values)

Diet	Replication	Initial		Final		Net gain		Growth (%)	
		Length	Weight	Length	Weight	Length	Weight	Length	Weight
CM	1	3.4	0.203	7.08	1.957	3.680	1.754	108.24	864.04
	2	3.1	0.190	7.30	1.907	4.20	1.717	135.48	903.68
	3	3.6	0.233	7.20	2.479	3.6	2.246	100.00	963.95
	4	3.0	0.159	7.10	2.581	4.1	2.422	136.67	1523.27
		(3.28)	(0.196)	(7.17)	(2.234)	(3.89)	(2.035)	(120.096)	(1063.74)
PM	1	3.3	0.175	6.0	1.172	2.7	0.997	81.82	569.71
	2	3.6	0.230	5.99	1.344	2.39	1.114	66.39	484.35
	3	3.7	0.235	6.10	1.194	2.40	0.959	64.86	408.09
	4	3.0	0.187	6.05	1.385	3.05	1.198	101.66	640.64
		(3.4)	(0.207)	(6.04)	(1.274)	(2.635)	(1.049)	(78.86)	(525.69)
SW	1	3.5	0.215	6.85	1.626	3.35	1.411	95.71	656.28
	2	3.4	0.213	6.90	1.730	3.50	1.517	102.94	712.206
	3	3.5	0.219	6.80	1.872	3.30	1.653	94.29	754.79
	4	3.1	0.180	6.75	1.498	3.65	1.318	117.75	732.22
		(3.38)	(0.207)	(6.83)	(1.682)	(3.45)	(1.475)	(102.67)	(713.88)
SM	1	3.0	0.191	5.30	0.920	2.30	0.729	76.67	381.67
	2	3.4	0.217	4.90	0.742	1.50	0.525	44.11	241.94
	3	3.0	0.169	6.20	0.718	3.20	0.549	106.67	324.85
	4	2.9	0.164	5.0	1.043	2.10	0.879	72.41	535.97
		(3.08)	(0.185)	(5.35)	(0.856)	(2.275)	(0.671)	(74.97)	(371.11)
MY	1	3.0	0.171	5.95	1.212	2.95	1.041	98.33	608.77
	2	3.1	0.189	6.45	1.423	3.35	1.234	108.06	652.91
	3	3.0	0.180	6.25	1.110	3.25	0.930	108.33	516.67
	4	3.1	0.195	6.25	1.184	3.15	0.989	101.61	507.18
		(3.05)	(0.184)	(6.23)	(1.232)	(3.18)	(1.067)	(104.09)	(571.38)

ANOVA TABLE 2

Analysis of variance of the data on gain in length and weight of *P. monodon* juveniles fed on diets based on different protein sources.

Source of variation	Degrees of freedom	Sum of squares	Mean sum of squares	F Value
Gain in length:				
Between diets	4	6.662	1.656	11.265**
Error	15	2.210	0.147	
Total	19	8.832		

Standard error of treatment means = 0.271

Critical difference = 0.799

Comparison of means based on critical difference

Diets	CM	SW	MY	PM	SM
Ranked means (cm)	3.89	3.45	3.18	2.65	2.28

Gain in weight				
Between diets	4	4.304	1.076	26.589**
Error	15	0.607	0.040	
Total	19	4.911		

Standard error of treatment means = 0.141

Critical difference = 0.417

Diets	CM	SW	MY	PM	SM
Ranked means (g)	2.035	1.475	1.069	1.047	0.671

Underscored means are not significantly different

** Significant at 1% level.

Table 9. Specific growth rate (%) of P. monodon juveniles fed on diets based on different protein sources

Diets	Replication	Average initial weight (g)	Average final weight (g)	Specific growth rate (%)	Mean
CM	1	0.203	1.957	5.395	5.78
	2	0.190	1.907	5.491	
	3	0.233	2.479	5.630	
	4	0.159	2.581	6.635	
PM	1	0.175	1.172	4.527	4.34
	2	0.230	1.344	4.203	
	3	0.235	1.194	3.870	
	4	0.187	1.385	4.767	
SW	1	0.215	1.626	4.817	4.99
	2	0.213	1.730	4.987	
	3	0.219	1.872	5.109	
	4	0.180	1.498	5.045	
SM	1	0.191	0.920	3.743	3.63
	2	0.217	0.742	2.927	
	3	0.169	0.718	3.444	
	4	0.164	1.043	4.405	
MY	1	0.171	1.212	4.663	4.53
	2	0.189	1.423	4.807	
	3	0.180	1.110	4.331	
	4	0.195	1.184	4.294	

ANOVA TABLE 3

Analysis of variance of the data on specific growth rate of P. monodon juveniles fed on diets based on different protein sources.

Source of variation	Degrees of freedom	Sum of squares	Mean sum of squares	F Value
Between diets	4	17.926	4.482	11.72**
Error	15	5.732	0.382	
Total	19	23.658		

Standard error of transformed treatment means = 0.437

Critical difference = 1.288

Comparison of transformed means based on critical difference

Diets	CM	SW	MY	PM	SM
Transformed mean values	13.91	12.91	12.29	12.02	10.98
	———	———			
		———	———	———	
			———	———	———

Underscored means are not significantly different

** Significant at 1% level

Plate IV

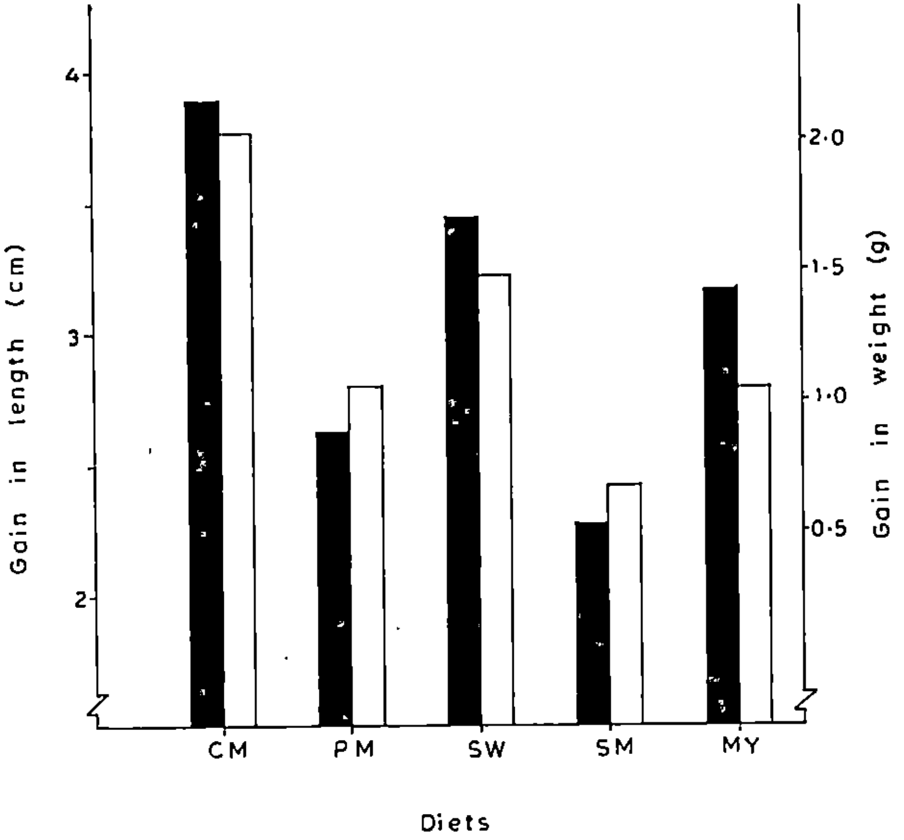

Fig. 5

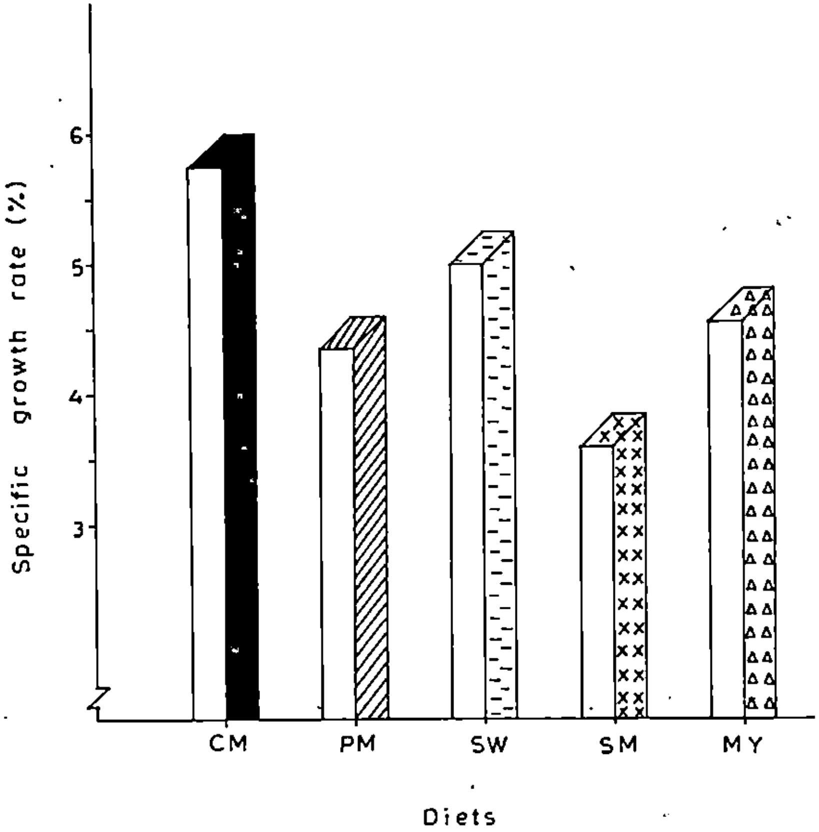

Fig. 6

protein sources (ANOVA Table 4). Diets from marine yeast (MY) and soybean meal (SM) gave relatively higher values (3.26 and 3.12) than those fed with diets from animal protein sources. The FCR obtained for diets from different animal protein sources did not vary markedly and ranged from 2.18 for clam meal (CM) to 3.06 for prawn meal.

4.5.2.5 Protein Efficiency Ratio

Protein efficiency ratios recorded for different treatments showed significant variations ($P < 0.01$) and are given in Table 11, Fig. 7, ANOVA Table 5). Diets formulated from animal protein sources recorded high PER values compared to plant or single cell protein sources, the highest PER of 1.13 was recorded for clam meal (CM) followed by slaughter house waste meal (SM) where the PER was 0.96. However among the different diets, the lowest PER of 0.77 was recorded for marine yeast (MY).

4.5.2.6 Protein Digestability

Table 12 and Fig. 8 give the results of the experiment conducted to determine the protein digestability coefficient of the five formulated feeds to the prawns. Analysis of variance of the data showed significant variation ($P < 0.01$) between treatments (ANOVA Table 6). The protein digestability coefficient was maximum (92.22%) with pellet CM followed by the pellet SW (88.52%). The least protein digestability was observed in the case of pellet SM (84.39%) where as the pellets PM and MY recorded protein digestability of 86.54 and 86.18% respectively.

Table 10. Food conversion ratio of *P. monodon* juveniles fed on diets based on different protein sources

Diet	Replication	Average initial weight (g)	Average final weight (g)	Average live weight gain (g)	Av. wt. of food consumed (g)	Food conversion ratio	Mean
CM	1	0.203	1.957	1.754	3.596	2.05	2.18
	2	0.190	1.907	1.717	3.769	2.10	
	3	0.233	2.479	2.246	5.211	2.32	
	4	0.159	2.581	2.422	5.449	2.25	
PM	1	0.175	1.172	0.997	3.141	3.15	3.06
	2	0.230	1.344	1.114	3.398	3.05	
	3	0.235	1.194	0.959	2.829	2.95	
	4	0.187	1.385	1.198	3.702	3.09	
SW	1	0.215	1.626	1.411	3.767	2.67	2.59
	2	0.213	1.730	1.518	3.825	2.52	
	3	0.219	1.872	1.653	4.116	2.49	
	4	0.180	1.498	1.318	3.559	2.70	
SM	1	0.191	0.920	0.729	2.275	3.12	3.12
	2	0.217	0.742	0.525	1.606	3.06	
	3	0.169	0.718	0.549	1.724	3.14	
	4	0.164	1.043	0.879	2.777	3.16	
MY	1	0.171	1.212	1.041	3.539	3.40	3.26
	2	0.189	1.423	1.234	4.134	3.35	
	3	0.180	1.110	0.930	2.928	3.12	
	4	0.195	1.184	0.989	3.165	3.20	

ANOVA TABLE 4

Analysis of variance of the data on food conversion ratio of *P. monodon* juveniles fed on diets based on different protein sources.

Source of variation	Degrees of freedom	Sum of squares	Mean sum of squares	F Value
Between diets	4	10.096	2.524	76.48**
Error	15	0.498	0.033	
Total	19	10.594		

Standard error of transformed treatment means = 0.128

Critical difference = 0.378

Comparison of transformed means based on critical difference

Diets	MY	SM	PM	SW	CM
Transformed mean values	10.40	10.17	10.07	9.26	8.49

Underscored means are not significantly different

** Significant at 1% level.

Table 11. Protein efficiency ratio of *P. monodon* juveniles fed on diets based on different protein sources

Diets	Replication	Average initial weight (g)	Average final weight (g)	Average live wt. gain (g)	Av. wt. of food consumed (g)	Average protein consumed	Protein efficiency ratio	Mean
CM	1	0.203	1.957	1.754	3.596	1.447	1.212	1.13
	2	0.190	1.907	1.717	3.769	1.517	1.132	
	3	0.233	2.479	2.246	5.211	2.097	1.071	
	4	0.159	2.581	2.422	5.449	2.193	1.105	
PM	1	0.175	1.172	0.997	3.141	1.257	0.793	0.817
	2	0.230	1.344	1.114	3.398	1.360	0.819	
	3	0.235	1.194	0.959	2.829	1.132	0.847	
	4	0.187	1.385	1.198	3.702	1.481	0.809	
SW	1	0.215	1.626	1.411	3.767	1.514	0.932	0.96
	2	0.213	1.730	1.518	3.825	1.538	0.987	
	3	0.219	1.872	1.653	4.116	1.655	0.999	
	4	0.180	1.498	1.318	3.559	1.431	0.922	
SM	1	0.191	0.920	0.729	2.275	0.941	0.798	0.79
	2	0.217	0.742	0.525	1.606	0.645	0.814	
	3	0.169	0.718	0.549	1.724	0.693	0.793	
	4	0.164	1.043	0.879	2.777	1.116	0.788	
MY	1	0.171	1.212	1.041	3.539	1.407	0.740	0.769
	2	0.189	1.423	1.234	4.134	1.644	0.751	
	3	0.180	1.110	0.930	2.928	1.164	0.799	
	4	0.195	1.184	0.989	3.165	1.258	0.786	

ANOVA TABLE 5

Analysis of variance of the data on protein efficiency ratio of *P. monodon* juveniles fed on diets based on different protein sources.

Source of variation	Degrees of freedom	Sum of squares	Mean sum of squares	F Value
Between diets	4	3.184	0.796	73.70**
Error	15	0.162	0.0108	
Total	19	3.346		

Standard error of transformed treatment means = 0.073

Critical difference = 0.216

Comparison of transformed means based on critical difference

Diets	CM	SW	PM	SM	MY
Transformed mean values	6.101	5.621	5.186	5.126	5.030
			______	______	______

Underscored means are not significantly different

** Significant at 1% level.

Table 12. **Protein digestability coefficient of the formulated feeds fed to *P. monodon* juveniles for a period of 20 days**

Treatments	* Food consumed	* Excreta produced	* Protein in feed	* Protein in excreta	* Protein digested (p)	* Protein ingested (q)	* Protein digestability coefficient ± SD (p/q)
CM	2.143	0.272	40.24	24.60	0.795	0.862	92.22 ± 0.54
PM	1.563	0.386	40.61	22.76	0.547	0.635	86.18 ± 0.21
SW	1.815	0.328	40.30	25.59	0.647	0.731	88.52 ± 0.44
SM	0.995	0.336	40.17	18.55	0.337	0.399	84.39 ± 0.49
MY	1.636	0.433	39.76	20.30	0.563	0.651	86.54 ± 0.22

* Average of four values

ANOVA TABLE 6

Analysis of variance of the data on protein digestability coefficient of formulated feeds fed to P. monodon juveniles.

Source of variation	Degrees of freedom	Sum of squares	Mean sum of squares	F Value
Between diets	4	117.809	29.45	20.71**
Error	15	2.191	0.146	
Total	19	120.00		

Standard error of transformed treatment means = 0.270

Critical difference = 0.796

Comparison of transformed means based on critical difference

Diets	CM	SW	MY	PM	SM
Transformed mean values	73.805	70.198	68.48	68.183	66.733

Underscored means are not significantly different

** Significant at 1% level.

Plate V

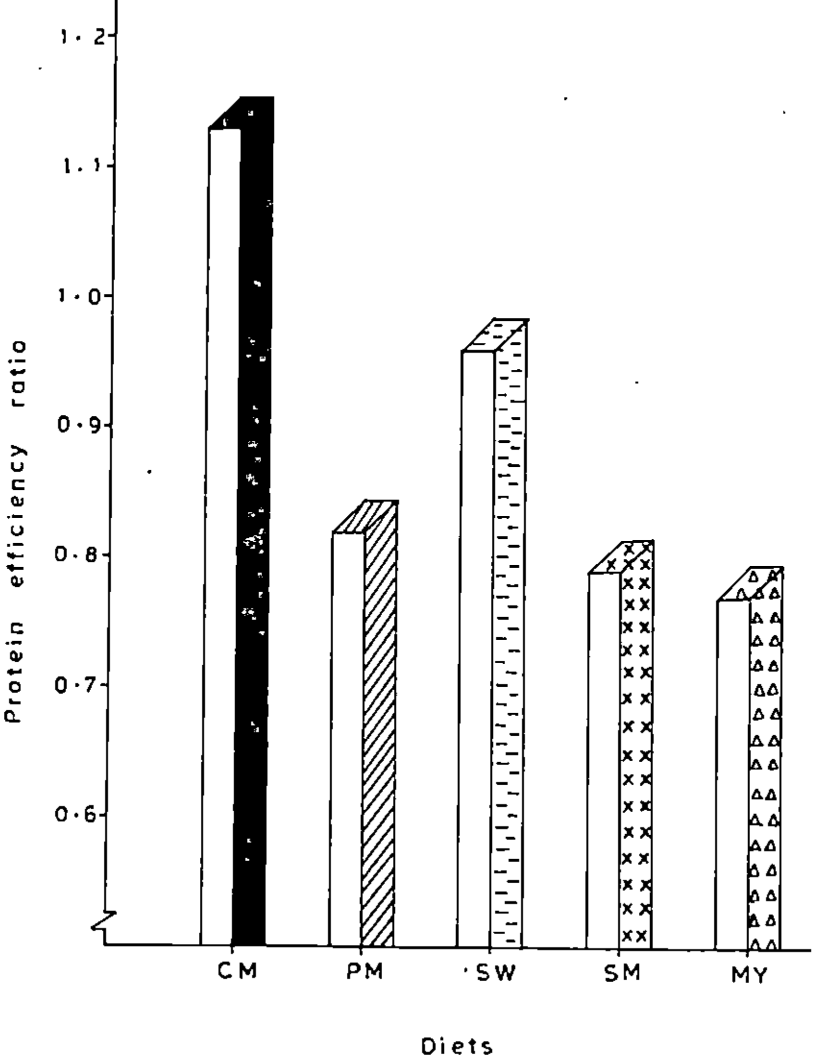

Fig. 7

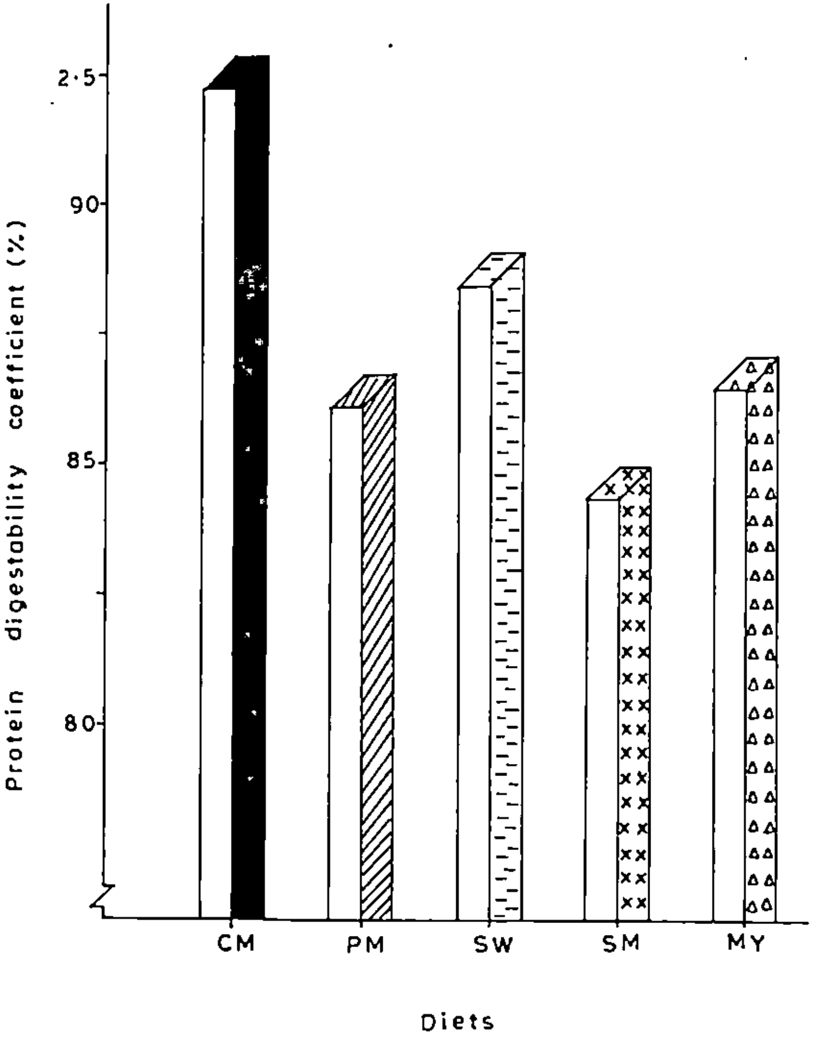

Fig. 8

4.5.3 Biochemical Composition of the Prawn

With a view to ascertaining whether there is any significant influence of the diets from various protein sources, on the body composition, biochemical composition of the whole prawn was evaluated after the experiments. The percentages of moisture, ash, protein, lipids and carbohydrates in prawns from different dietary treatments are shown in Table 13, Fig. 9.

Analysis of variance of the data shows that protein sources used in the diets significantly influence ($P < 0.05$) the moisture, protein, fat and ash contents of the prawns (ANOVA Table 8). The moisture content of prawns fed on diets with plant protein sources was relatively higher than those fed with animal protein sources or single cell protein sources. However there was no significant difference between prawns fed on diets containing the various animal protein sources, as their moisture content ranged from 75.16% to 76.58%. The highest moisture content was recorded in prawns fed on soybean meal (SM) (77.32%). Prawns fed with marine yeast (MY) recorded a moisture content of 75.62%.

Protein contents of prawns showed distinct variation in relation to the dietary protein sources. Diet based on plant protein source produced significantly lower protein content ($P < 0.05$) than those based on animal on single cell protein source. Among the different treatments with animal protein sources, the protein contents of the prawns fed on diet from slaughter house waste (SW) was 66.37%, and that from clam meal (CM) was 65.71%, which were relatively higher than those obtained for prawns fed on prawn meal (PM) whose protein content was 64.22% only. The prawns fed on diets with soybean meal (SM) had the lowest protein content (59.86%).

It was observed that prawns fed on diets containing animal protein sources had higher fat contents (15.53% to 16.9%), when compared to prawn fed on diet with plant protein source or single cell protein source (14.88% and 15.19%). Prawns fed on diet from slaughter house waste meal had the highest fat content (16.9%) whereas the prawns fed on diet containing soybean meal had the lowest fat content (14.88%). Differences between the fat content of prawns fed on diets with clam meal (16.10%), prawn meal (15.53%) and marine yeast (15.19%) were insignificant. Total ash content was relatively higher in prawns fed on diets with plant protein source and single cell protein source than those fed with animal protein sources. Highest ash content was recorded in prawns fed with soybean meal (22.82%) followed by marine yeast (19.48%). Among the animal protein sources, prawns fed on prawn meal based diet had the highest ash content (18.12%) whereas prawns fed on diets with slaughter house waste (SW) had the lowest ash content (15.02%).

The calcium content in prawns fed on diets with plant protein sources is relatively lower than those fed on animal or single cell protein sources. Among the diets containing animal protein sources the highest calcium content was recorded in prawns fed with slaughter house waste (4.20%) and the lowest in diet containing clam meal (3.75%). In the case of prawns fed with diet based on marine yeast, the calcium content was 4.05% and those fed on soybean meal based diet recorded 3.70%.

Phosphorus content of prawns fed with soybean meal based diet was 0.59% which was significantly lower ($P < 0.05$) than those fed with other

Table 13. Biochemical composition of *P. monodon* fed with diets based on different protein sources

Parameter* (%)	Treatments (Mean values)				
	CM	PM	SW	SM	MY
Moisture	76.23	76.58	75.16	77.32	75.62
Protein	65.71	64.22	66.37	59.86	63.31
Fat	16.10	15.53	16.9	14.88	15.19
Ash	16.30	18.12	15.02	22.82	19.48
Carbohydrate	1.89	2.13	1.71	2.44	2.02
Phosphorus	1.15	1.02	0.88	0.59	1.09
Calcium	3.75	4.20	3.96	3.70	4.05

* Average of four values.

Plate VI

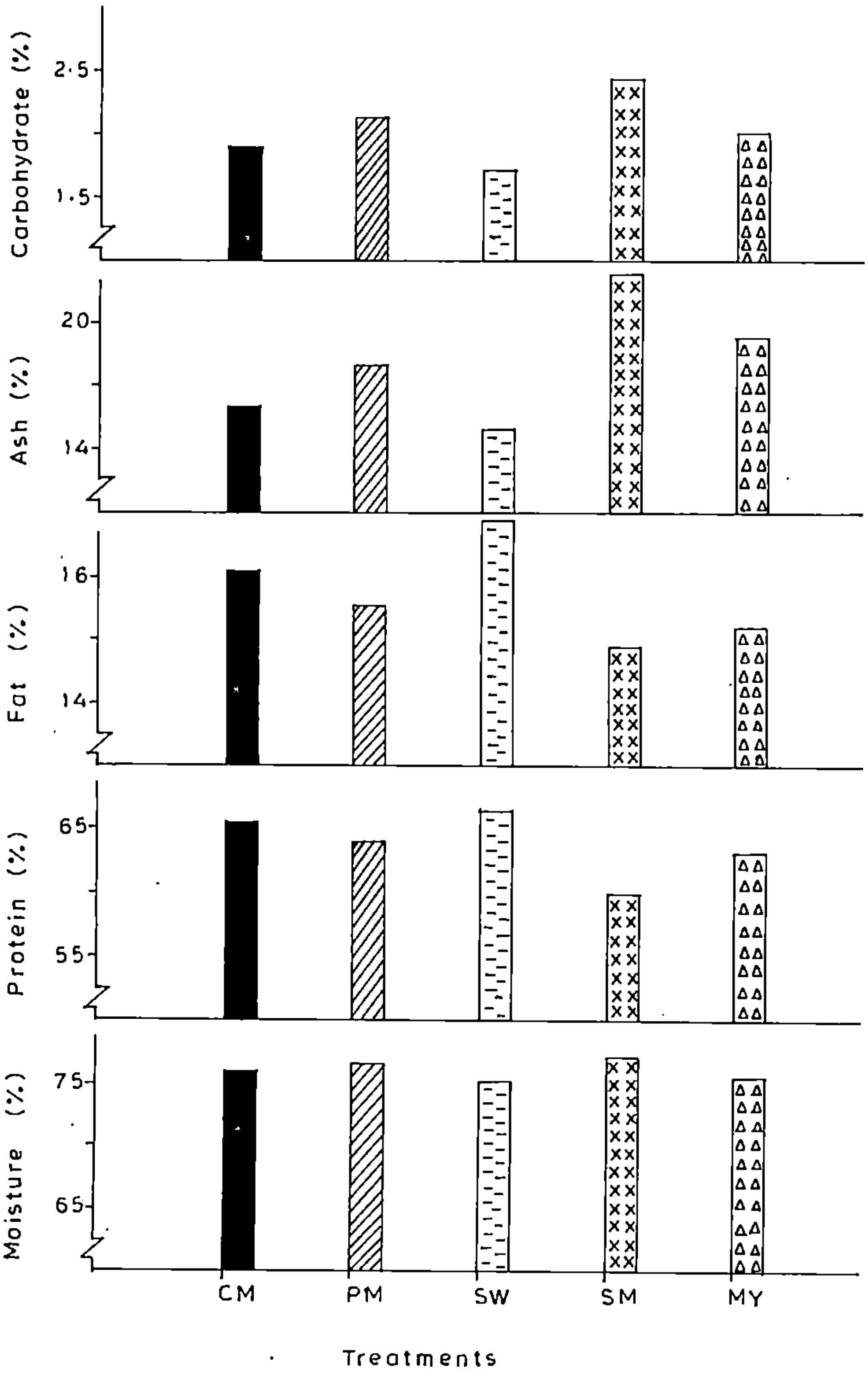

Fig. 9

ANOVA TABLE 7

Analysis of variance of the data on biochemical composition of *P. monodon* juveniles fed with diets based on different protein sources.

Source of variation	Degrees of freedom	Sum of squares	Mean sum of squares	F Value
Moisture				
Between treatments	4	8.978	2.2445	6.868**
Error	15	4.902	0.3268	
Total	19	13.88		

Standard error of transformed treatment means = 0.4042

Critical difference = 0.8614

Comparison of transformed mean based on critical difference

Treatment codes	SM	PM	CM	MY	SW
Transformed mean values	61.46	60.47	60.41	59.87	59.47

Protein

Source of variation	Degrees of freedom	Sum of squares	Mean sum of squares	F Value
Between treatments	4	36.899	9.225	15.566**
Error	15	8.889	0.5926	
Total	19	45.788		

Standard error of transformed treatment means = 0.544

Critical difference = 1.159

Treatment codes	SW	CM	PM	MY	SM
Transformed mean values	54.56	56.16	53.26	52.74	50.69

Fat

Between treatments	4	6.269	1.567	11.782**
Error	15	1.995	0.133	
Total	19	8.264		

Standard error of transformed treatment means = 0.258

Critical difference = 0.549

Treatment codes	SW	CM	PM	MY	SM
Tranformed mean values	24.27	23.66	23.21	22.94	22.69

Carbohydrate

Between treatments	4	5.448	1.362	2.961 [N.S]
Error	15	6.900	0.46	
Total	19	12.348		

Ash

Between treatments	4	102.24	25.560	9.598**
Error	15	39.95	2.63	
Total	19	142.19		

Standard error of transformed treatment means = 1.154

Critical difference = 2.459

Treatment codes	SM	MY	PM	CM	SW
Transformed mean values	28.54	26.19	25.19	23.81	22.80

Underscored means are not significantly different

** Significant at 5% level

N.S Not significant at 5% level.

dietary protein sources, in which the phosphorus content varied between 0.88 and 1.15%. Among the diets based on animal protein sources, the phosphorus content was relatively higher in prawns fed on diet containing clam meal (1.15%), while lower in those fed on diet containing soybean meal (0.59%). In the prawns fed on the diet containing marine yeast and slaughter house waste meal the phosphorus content was 1.09 and 0.88% respectively.

4.5.4 Oxygen Consumption, Ammonia Excretion and O : N Ratios

Oxygen consumption and ammonia excretion rates of the test prawns (Table 14, Fig. 10) were significantly influenced ($P < 0.01$) by the protein sources (ANOVA Table 8). The oxygen consumption rate of prawns fed on diet with animal protein sources was higher than those fed on diet with single cell protein source. But the rate was not significantly different from that of prawns fed on plant protein (soybean meal) source. Among the animal protein sources highest oxygen consumption rate was observed in prawns fed on prawn meal (0.173 mg O_2/g/hr), and minimum in prawns fed on diet containing slaughter house waste (0.143 mg O_2/g/hr).

The oxygen consumption rate of prawns fed on diet containing soybean meal and marine yeast was 0.172 mg O_2/g/hr and 0.149 mg O_2/g/hr respectively.

The prawns fed on the diets with animal protein sources excreted less ammonia when compared to those fed on diets with plant or single cell protein sources. The lowest rate of ammonia excretion of 0.0157 mg NH_3-N/g/hr was recorded in prawns fed on clam meal based diet and

Table 14. Oxygen consumption, ammonia excretion rate and O : N ratios of *P. monodon* juveniles fed with diets based on different protein sources

Parameters *	DIETS (Mean ± SD)				
	CM	PM	SW	SM	MY
Length (cms)	5.82 ± 0.19	5.74 ± 0.29	5.80 ± 0.20	5.70 ± 0.22	5.78 ± 0.30
Weight (g)	1.352 ± 0.17	1.350 ± 0.23	1.352 ± 0.21	1.346 ± 0.16	1.350 ± 0.12
Oxygen consumption rate - metabolic rate (mgO_2/g/hr)	0.1705 ± 0.0114	0.173 ± 0.004	0.143 ± 0.001	0.172 ± 0.002	0.149 ± 0.005
Ammonia excretion rate (mgNH_3-N/g/hr)	0.0157 ± 0.001	0.0183 ± 0.001	0.0192 ± 0.001	0.0285 ± 0.001	0.0234 ± 0.001
O : N ratio	9.52	8.32	6.53	5.27	5.60

* Average of four values

Plate VII

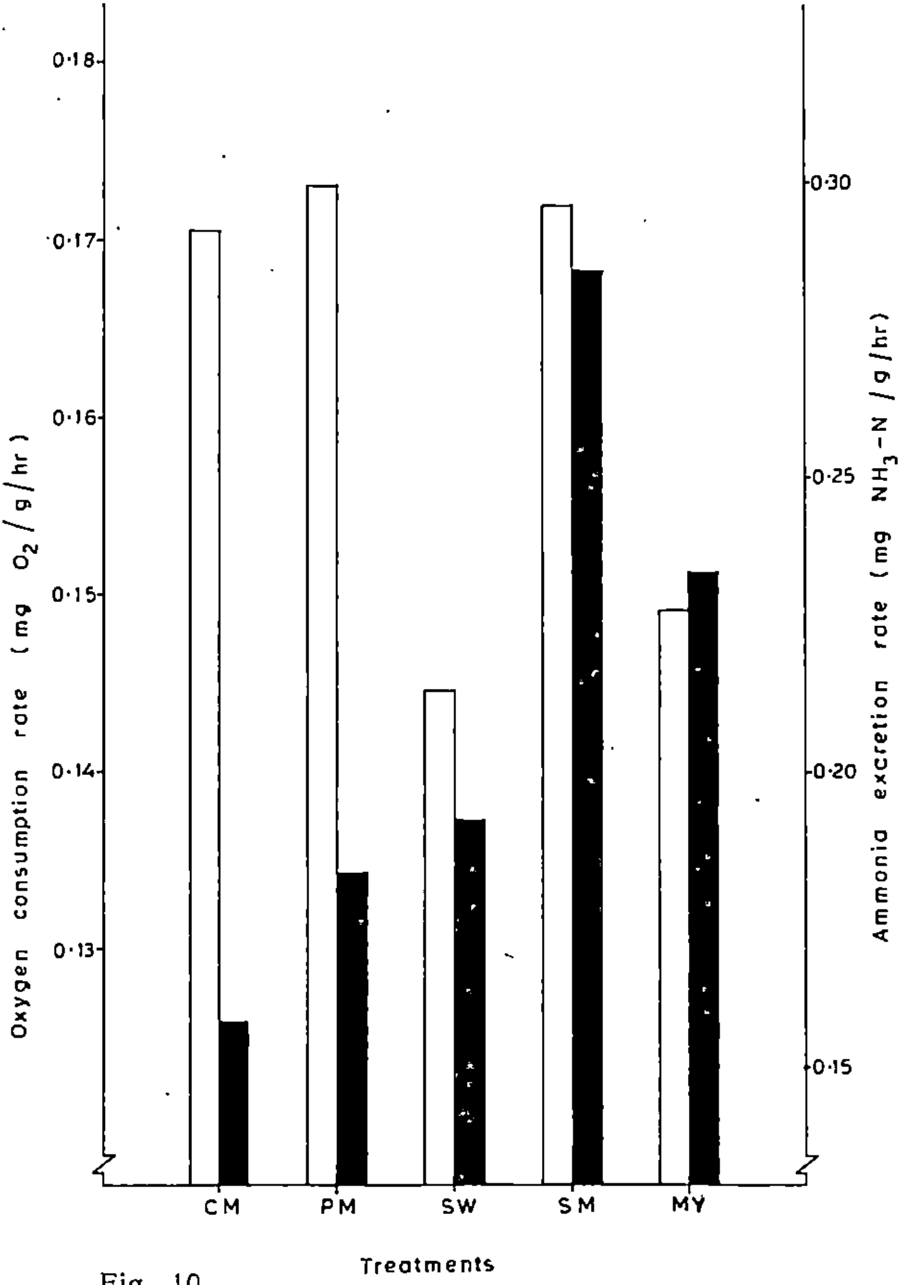

Fig. 10

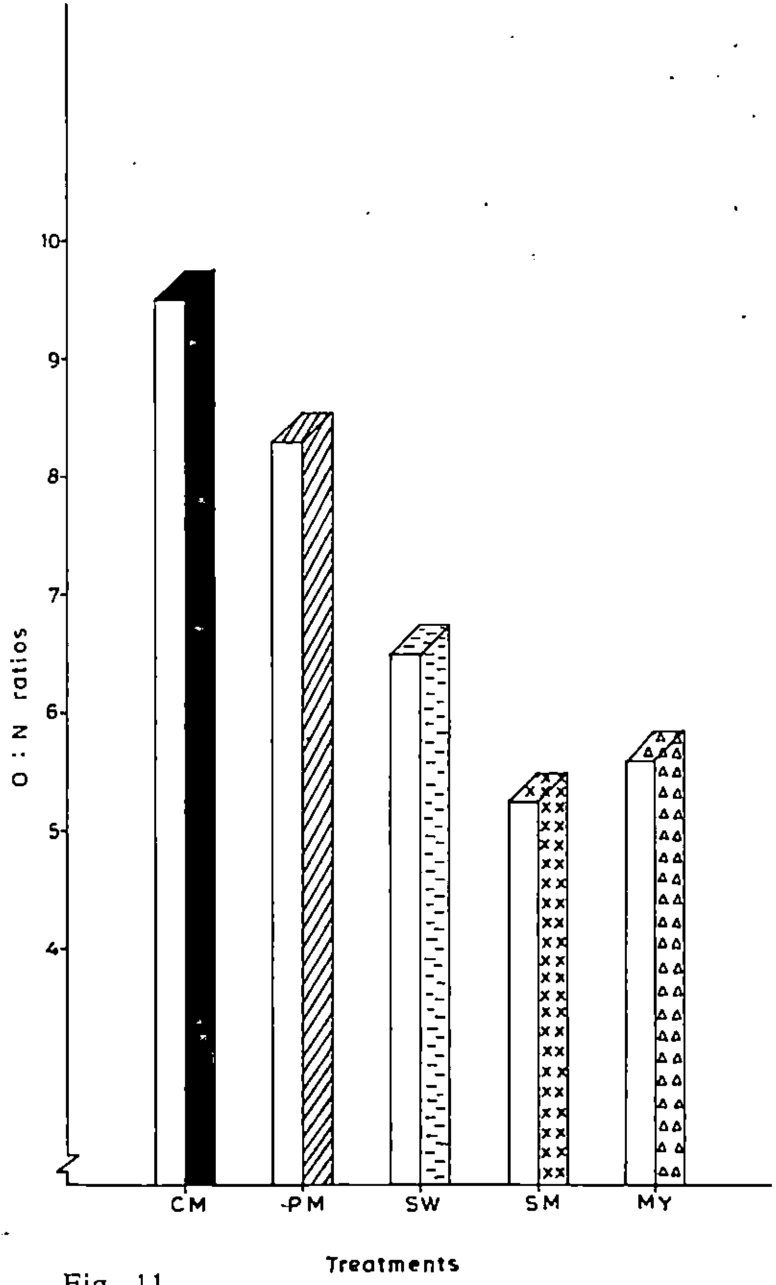

Fig. 11

ANOVA TABLE 8

Analysis of variance of the data on oxygen consumption, ammonia excretion and O : N ratios of *P. monodon* juveniles fed with diets based on different protein sources.

Source of variation	Degrees of freedom	Sum of squares	Mean sum of squares	F Value
Oxygen consumption rate				
Between treatments	4	0.163	0.040	22.63**
Error	15	0.027	0.0018	
Total	19	0.190		

Standard error of transformed treatment means = 0.03

Critical difference = 0.088

Comparison of transformed treatment means based on critical difference

Treatments	PM	SM	CM	MY	SW
Transformed means value	2.383	2.376	2.366	2.212	2.167

Ammonia excretion rate

Between treatments	4	0.151	0.0377	62.91**
Error	15	0.009	0.0006	
Total	19	0.160		

Standard error of transformed treatment means = 0.055

Critical difference = 0.161

Treatments	SM	MY	SW	PM	CM
Transformed means value	0.967	0.876	0.794	0.775	0.718

O : N ratios

Between treatments	4	64.983	16.245	486.39**
Error	15	0.501	0.0334	
Total	19	65.484		

Standard error of transformed treatments means = 0.129

Critical difference = 0.381

Treatments	CM	PM	SW	MY	SM
Transformed means value	17.971	16.76	14.806	13.699	13.271

Underscored means are not significantly different

** Significant at 1% level.

highest rate 0.0285 mg NH_3-N/g/hr) in prawns fed on diet containing soybean meal. Ammonia excretion rates observed in prawns fed on diets containing marine yeast and slaughter house were 0.234 and 0.0192 mg $NH_3{}^{-N}$/g/hr respectively.

O : N ratios (Fig. 11) were also significantly influenced ($P < 0.01$) by the dietary protein source (ANOVA Table 9). O : N ratios of prawns fed on diet with animal protein sources are higher than those fed on plant on single cell protein source. The highest O : N ratio (9.52) was recorded in prawns fed on clam meal based diet and the lowest (5.27) in prawns fed on soybean meal based diet. The O : N ratios obtained in prawns fed on diet containing marine yeast and slaughter house waste were 5.6 and 6.53 respectively.

4.6 Study on Optimum Food Ration

Table 15 presents the data on the effect of food rations on food consumption and growth of P. monodon juveniles. The physico-chemical conditions of water during the experimental period are given in Table 16.

The analysis of variance of the data shows that different food rations significantly influence ($P < 0.01$) the growth rate of test animals (ANOVA Table 19), the maximum live weight gain, being seen in prawns fed on food ration at the rates of 15% of their body weight. A decrease in live weight and low survival rate (32%) was observed in prawns reared without food. Gain in weight and survival rate increase with increase in food ration until the ration reached 15% of the body weight, after which the gain in weight decreases. The food intake is also low beyond 15% food ration.

Table 15. Effect of food ration on food consumption and growth of *P. monodon* juveniles

Parameters*	0	3	6	9	12	15	18	21
Mean initial weight (g)	0.236	0.213	0.201	0.208	0.188	0.208	0.205	0.220
Mean weight after 3 weeks (g)	0.141	0.277	0.391	0.539	0.543	0.632	0.627	0.579
Mean live weight gain (g)	-0.095	0.064	0.190	0.331	0.355	0.424	0.422	0.359
Total food consumption per animal (g)	-	0.141	0.389	0.745	0.905	1.230	1.413	1.400
Food conversion ratio	-	2.20	2.05	2.25	2.55	2.90	3.35	3.90
Specific growth rate (g)	-1.805	1.251	3.16	4.531	5.04	5.29	5.097	4.608
Survival (%)	32	56	80	82	86	92	90	86

* Average of three values.

Table 16. Water quality parameters of the experimental tanks recorded during the study on effect of food ration

	Water temperature (°C)	Salinity (ppt)	pH	Dissolved oxygen (ppm)
Mean ± SD	27.3 ± 0.64	20.5 ± 0.46	8.13 ± 0.21	6.38 ± 0.74
Range	25.50 - 28.50	20.0 - 21.2	7.97 - 8.49	5.20 - 7.01

ANOVA TABLE 9

Analysis of variance of the data on gain in weight of *P. monodon* juveniles obtained during the study on the effect of food ration.

Source of variation	Degrees of freedom	Sum of squares	Mean sum of squares	F Value
Between treatment	7	0.5420	0.0774	66.15**
Error	16	0.0187	0.0017	
Total	23	0.5607		

Standard error of treatment means = 0.028

Critical difference = 0.082

Comparison of treatment means based on critical difference

Food rations (% of the body wt)	15	18	21	12	9	6	3	0
Ranked means	0.424	0.422	0.359	0.355	0.331	0.190	0.064	-0.095
	———	———	———	———				
			———	———	———			

Underscored means are not significantly different

** Significant at 1% level.

Plate VIII

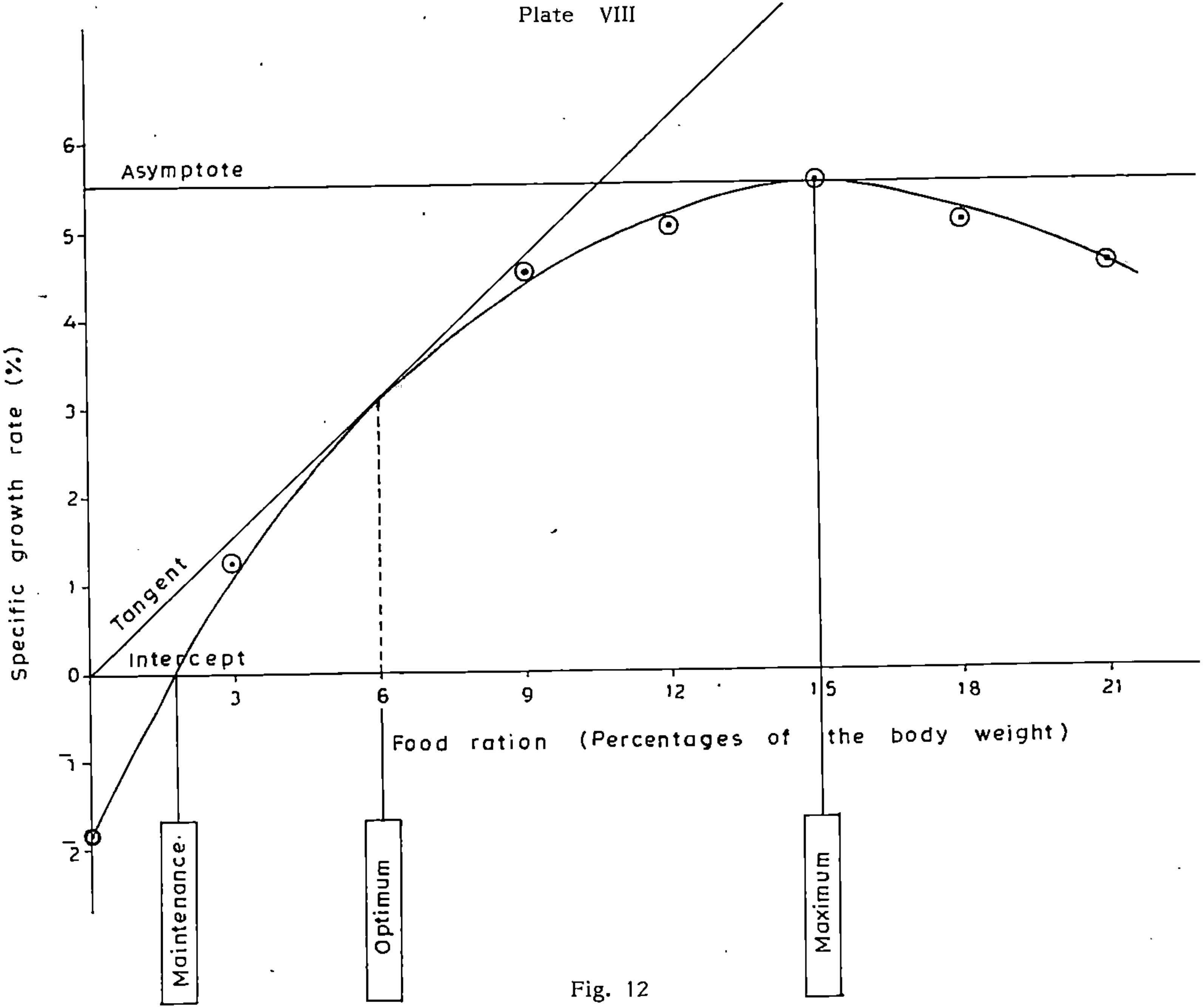

Fig. 12

The graph (Fig. 12) shows that maintenance ration is at 1.8% of the body weight while optimum and maximum rations are at 6.0% and 15% respectively.

4.7 Study on Optimum Feeding Frequency

Data on the effect of feeding frequency on food consumption, growth and survival in juveniles of *P. monodon* are shown in Table 17. Water quality parameters of the experimental tanks are presented in the Table 18.

Food consumption, growth and survival increased as the feeding frequency was raised. However, food consumption and growth increased only upto an increase of 3 meal/day and there after these parameters show a declining trend (Fig. 13). There was an increase of 46% and 31% in food consumption and gain in weight as the feeding frequency was raised from once a day to thrice a day respectively. Analysis of variance of the data shows that frequency of feeding significantly influence ($P < 0.01$) the growth of test animals (ANOVA Table 10). The highest live weight gain was recorded in the case of prawns, which were fed thrice daily (4.45) and lowest in those fed once a day (3.37).

In the case of prawns fed once daily, feeding time was seen to influence food consumption, growth and survival. Thus the prawns fed during night (8 pm) gave a better result than those fed during day time. Again in twice a day meal frequency, appreciable growth was obtained when meal was offered during morning-night hours instead of morning - noon and noon - night hours (Table 17).

Table 17. Effect of feeding frequency on food consumption, growth and survival of *P. monodon* juveniles

Parameters*	Feeding frequency (number of meals/day)						
	Once a day		Twice a day			Thrice a day	4 times a day
	0800 hrs	2000 hrs	0800 hrs & 2000 hrs	0800 hrs & 1400 hrs	1400 hrs & 2000 hrs	0800,1400, 2000 hrs	0800, 1100, 1400, 1700, 2000 hrs.
	(A)	(B)	(C)	(D)	(E)	(F)	(G)
Mean initial weight (g)	0.240	0.201	0.198	0.216	0.224	0.202	0.220
Mean weight after 4 weeks (g)	0.617	0.636	0.667	0.668	0.670	0.702	0.718
Mean live weight gain (g)	0.377	0.435	0.469	0.452	0.456	0.494	0.498
Total food consumption per animal (g)	1.206	1.465	1.680	1.600	1.620	1.884	1.733
Specific growth rate (%)	3.37	4.11	4.34	4.03	3.91	4.45	4.22
Survival (%)	82	86	88	86	84	92	90

* Average of 3 values.

Table 18. Water quality parameters of the experimental tanks recorded during feeding frequency study

	Water temperature (°C)	Salinity (ppt)	pH	Dissolved Oxygen (ppm)
Mean ± SD	26.9 ± 1.40	20.0 ± 0.5	7.95 ± 0.27	5.64 ± 0.65
Range	25.5 - 29.8	19.5 - 21.2	7.63 - 8.37	4.80 - 6.45

ANOVA TABLE 10

Analysis of variance of the data on gain in weight of *P. monodon* juveniles obtained during the study on the effect of feeding frequency.

Source of variation	Degrees of freedom	Sum of squares	Mean sum of squares	F Value
Between treatments	6	0.026	0.0043	6.143**
Error	14	0.010	0.0007	
Total	20	0.036		

Standard error of treatment means = 0.0216

Critical difference = 0.064

Comparison of treatment means based on critical difference

Feeding frequency	G	F	C	E	D	B	A
Ranked means (g)	0.498	0.494	0.469	0.456	0.452	0.435	0.377

Underscored means are not significantly different

** Significant at 1% level.

Plate IX

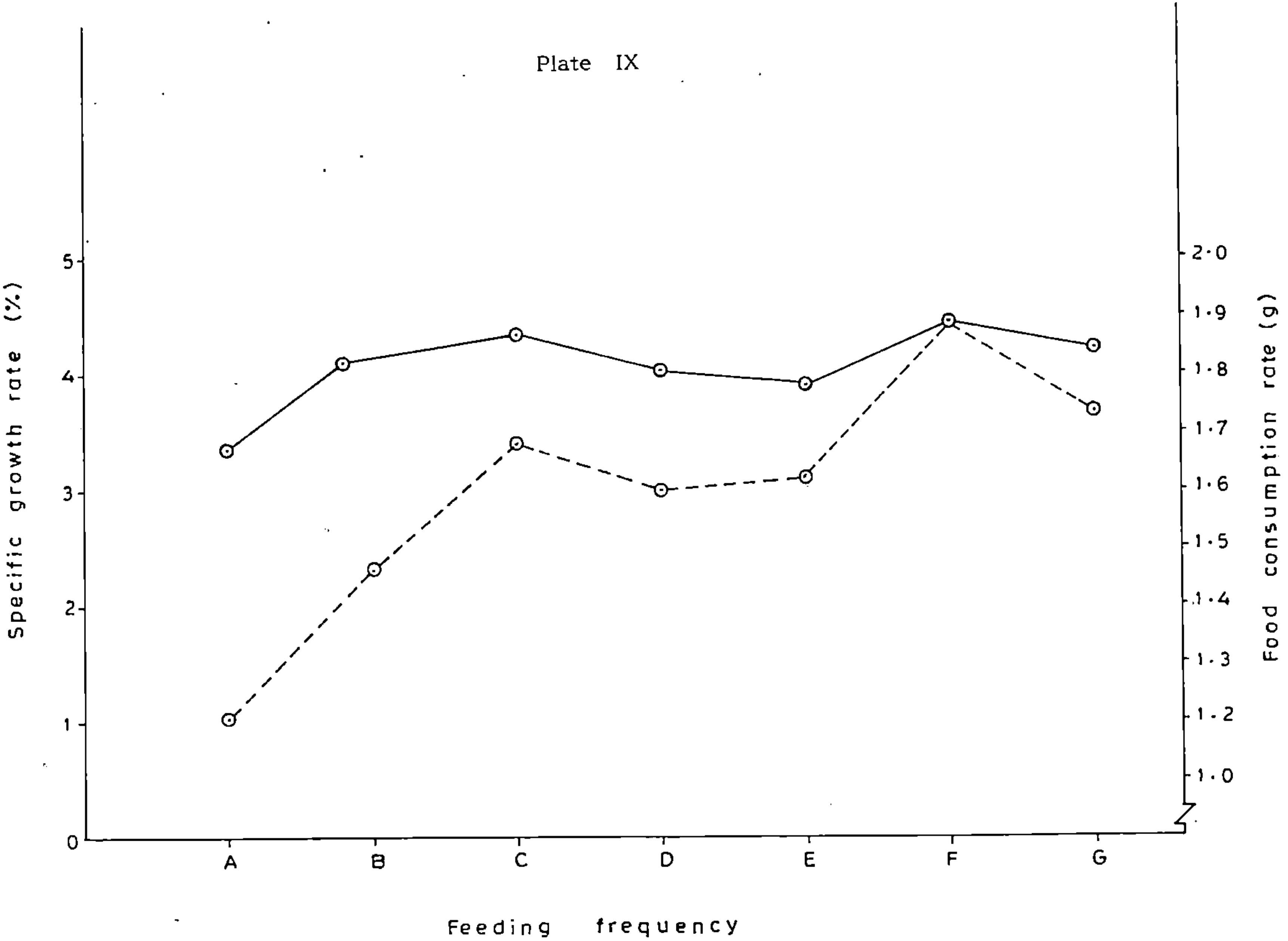

Fig. 13

V. DISCUSSION

V DISCUSSION

5.1 Keeping Quality of Formulated Feeds

The quality of the feed is of utmost importance in the formulation of a diet. A good quality feed is one which is able to maintain its nutritional status for a few weeks of storage. Fowler and Banks (1967) have found no alterations in the nutritional status of the feed pellets stored for few weeks, but storage for greater periods had deleterious effect on the growth performance of fish. The feeds employed in the present study showed satisfactory keeping quality in their nutrient content for a period of four months. According to Hilton et al. (1977), the shelf life of a processed feed is dependent on the type of processing, storage temperature and moisture content of the diet.

5.1.1 Moisture

All the feeds used in the present study showed increased moisture content at the end of four months storage, the percentage increase being different for different feeds. The pellet MY absorbed more moisture (35.64%) than other pellets. Due to the increase in moisture content, the pellets became heavier which was evidenced by their increased sinking rate. The uptake of moisture during storage is due to the presence of hygroscopic ingredients mainly starch (Hastings, 1971) and porosity of feeds (Jayaram and Shetty, 1980). Since the pellet MY absorbed more moisture compared to other pellets, it can be inferred that it contained high amount of hygroscopic materials and is also more porous than the remaining feeds. Higher absorption of moisture by the pellet MY might also be due to its lower fat content. Jayaram and Shetty (1981) reported that higher fat content hardens the particle, thereby preventing the entry of moisture into the pellet. The reduced amount of moisture absorption by the feed pellets CM, PM and SW can be attributed to the same reason. Srikanth (1986) reported that high crude fibre content in feed will help in increased moisture absorption during storage. But contrary to this the feeds tested in the present study, the pellet SM recorded the least percentage increase in moisture (15.27%) after storage period, eventhough the crude fibre content of the feed is higher than all other feeds (7.02%).

5.1.2 Proteins

The crude protein decreased in the different pellets used in the present study during storage, the percentage decrease for pellets CM, PM, SW, SM and MY being 8.08%, 7.40%, 8.98%, 6.47% and 10.56%, respectively. The decrease in crude protein, may be due to the increase in moisture level of feeds during storage. Jayaram and Shetty (1980) and Venugopal and Keshavanath (1984) have also reported similar observations in the feeds they used.

5.1.3 Fat

A marginal decrease in the crude fat content was noted in all the five types of pellets, since the fat content varies inversely with moisture. Its decrease may be attributed to the increase in moisture, which may facilitate lipid hydrolysis. Lipid autoxidation taking place during storage can also cause decrease in fat content. Jayaram and Shetty (1980), and Venugopal and Keshavanath (1984) also reported a decrease in crude fat which they attributed to an increase in moisture content during storage and also to the degree of unsaturated oil present in the diet. This may be one of the reasons for the decrease in fat content noticed in the present investigation. Pellet MY showed the maximum percentage decrease (18.07%) compared to other pellets and the rate of reduction of fat among the different feeds may be directly linked to their moisture content.

5.1.4 Ash, Crude fibre and Carbohydrate

The variations noted in the values of ash and crude fibre are mainly due to the moisture content in the feeds, associated with changes during storage. The differences in the carbohydrate content may be linked to the changes in other components in the feed during storage.

5.1.5 Total Heterotrophic bacteria and Fungi

The total bacterial count and fungal count increased in all feeds after storage. Increase in moisture content during storage may be the reason for increased growth of the microorganisms. Cho et al. (1985) reported that increased microbial load results in decreased palatability and nutritive value of the feed.

5.2 Water Stability of Pelleted Feeds

In an aquaculture system, stability of compounded pellets is of utmost importance, as retension of nutritive compounds in the pelleted feeds for longer time without disintegration, makes them more efficient as well as economical. The formulated pelleted feeds should remain stable for at least one hour, so that they become available to the aquatic organisms.

Ingredient composition, nature of ingredients, type of processing and moisture content are known to influence the feed stability (Hastings, 1971; Kainz, 1977). In the present study, the pellet SM was the most stable at the end of three hours compared to the other diets tested. The high water stability of this pellet, may be due to the high amount of carbohydrate content in it. According to Stivers (1971) and Jayaram and Shetty (1981) the degree of stability is dependant on the gelatinization of starch content of the feed during cooking. Hastings (1971) has stated that higher fat content affects geletinization thereby reducing the stability of the feed. The present study shows that the water stability of the different feeds is in the order SM > CM > SW > PM > MY whereas the fat content of the feeds is in the order SW > CM > PM > SM > MY and the moisture content is in the order MY > PM > SW > CM > SM which show an inverse relationship of water stability with moisture content rather than with fat content.

Prawns being demersal, feed by grasping the feed pellets with pincer like appendages and masticate externally (Forster, 1971; Zein-Eldin and Meyers, 1973). Hence pellets with high water stability are likely to show better results. All the five pelleted feeds tested during the present investigation showed satisfactory stability upto three hours and hence may be suitable as prawn feeds. Too much stability is also not desirable, because the nutrients in them become unavailable to organisms owing to their bound form in addition to the higher production cost (Balazs et al., 1973).

The stability of the feeds decreases during storage which may be due to increase in moisture absorption by the diets. The percentage decrease in the water stability of feeds stored for four months was least in SM

(2.22%), followed by CM (2.52%), PM (3.24%), SW (3.31%), and MY (4.281%).

5.3 Evaluation of Efficiencies of Natural Protein Sources

5.3.1 Feeding Experiment

In the present study few locally available protein sources were tested in relation to their nutritional quality for juveniles of *P. monodon* based on survival, growth, food conversion ratio, protein efficiency ratio, protein digestibility, body composition and O:N ratios. These parameters were also employed by few earlier authors to evaluate the biological value of protein sources in penaeids and palaemonid prawns (Balazs and Ross, 1976; New, 1976).

5.3.1.1 Growth Studies

Among the different feeds tested in the present study, the diet containing clam meal (CM) gave the highest growth performance, followed by the diet containing slaughter house waste (SW). The lowest growth performance was recorded with soybean meal based diet (SM). The diet containing marine yeast (MY) and prawn meal (PM) were found to be superior to the plant protein source tested.

Most of the plant proteins have been shown to yield poor growth rates in prawns, when used individually, excepting a few like soybean meal, wheat gluten, peanut meal etc (Kanazawa *et al.*, 1970; Sick and Andrews, 1973; Deshimaru and Shigueno, 1972; Balazs *et al.*, 1973). Improved growth rates produced by some of the plant protein sources have been attributed to their higher polysaccharide contents than to monosaccharides (Sick and

Andrews, 1973). These plant protein sources have been observed to promote superior growth rates when mixed with animal protein sources (Lee, 1970).

Sick and Andrews (1973) have shown that soyabean meal is superior to other plant protein sources such as groundnut oil cake, cotton seed etc. According to Deshimaru and Shigueno (1972) the similarity of aminoacid profile of the soybean meal with that of prawns may be the reason for its better performance than other plant protein sources. However, in comparison with diets based on animal protein sources, soybean meal based diet produced lesser growth rate. In the present study also soybean meal based diet gave the lowest growth performance compared to diets containing animal or single cell protein sources.

One of the earliest artificial diet of prawn P. japonicus was derived from that of silkworm pupae, chinook salmon and brine shrimp (Kanazawa et al., 1970). Subsequently, Shigueno et al. (1972) have shown that high levels of protein obtained from several protein sources such as squid meal, white fish meal, dried euphausids and active sludge are efficient feed ingredients. These studies have led to the identification of several animal protein sources that can be efficiently used for formulation of feeds for prawns. Among the animal protein sources, protein of marine origin is preferred to fresh water origin not only due to the similarity in their amino acid profile, but also due to the better composition of unsaturated fatty acids essential for prawns and higher ash content (Cowey and Sargent, 1972; Boghen and Castell, 1981). For evaluating the nutritional efficiency of the feeds, based on a standard scale, Phillips and Brockway (1956) have suggested that feeds with essential amino acid composition similar to the test animal would be the most suitable for diet preparation in fishes. Ogino (1963) has also supported this suggestion. Deshimaru and Shigueno (1972) while agrreing with Phillips and Brockway (1956) have suggested that the same standard can be applied for the preparation of artificial feed for *P. japonicus*. Fresh clam meat is conventionally used as a feed for prawns, both in the nursery rearing phase as well as in growout systems. Kanazawa *et al.* (1970) reported that the fresh diet of short necked clam *Tapes philippinarum* gave superior growth compared to other compounded diets in *P. japonicus*. Surya Narayanan and Alexander (1972) have reported that clam meat (*Villorita*) is a good source of protein and contains maximum number of free amino acids. Forster and Beard (1973) reported higher growth rate and survival in *Palaemon serratus* using fresh clam meat diet. In the present study, black clam *Villorita cyprinoides* has been used for preparing the diet (CM) for *P. monodon* which gave higher growth performance compared to other animal or plant protein sources tested. Ahamad Ali (1982a) has also reported higher growth and survival rate in juveniles of *P. indicus* with diet containing dry clam meat powder of *Villorita cyprinoides*. But Vellegas (1978) has shown that the growth and survival of *P. monodon* larvae fed with fresh *Tapes* was only next to the compounded diets. Colvin (1976) also reported poor performance of *P. indicus* fed on fresh clam meat as control diet.

Prawn meal has also been regarded as a suitable protein source for the formulation of feed for penaeid prawns. Sick and Andrews (1973) obtained higher growth and survival in *P. duorarum* fed on diet with shrimp meal. Similarity of the amino acid profile may have resulted in the better performance of juvenile prawns fed on prawn meal based diet (Deshimaru and

Shigueno, 1972). But contrary to this, Pascual and Destajo (1978) observed poor growth and survival of *P. monodon* post-larvae fed on a diet prepared exclusively from shrimp head meal, since according to them shrimp head meal cannot satisfy the nutritional demands of the prawn post-larvae. Simpson *et al.* (1981) have reported that chitin and calcium carbonate present in the exoskeleton of prawns may have deleterious effects on the overall nutritional quality of diet which in turn affect the growth and survival of the organism fed on this diet. In the present study also prawn meal based diet fed to *P. monodon* juveniles produced low survival and growth compared to diets based on other animal protein sources.

A variety of single cell protein sources including marine yeast have been used for rearing the larval stages of penaeid prawns (Applebaum, 1979; Watanabe *et al.*, 1983). Marine yeasts *Saccharomyces cerevasiae* and *Rhodotorula aurantica* were successfully used in the larval rearing of *P. monodon* producing higher survival rate than those reared with algae (Aujero *et al.*, 1984). Marine yeast was also incorporated in the diet of fishes during growing stages (Matty and Smith, 1978; Atack *et al.*, 1979). But studies in this direction are limited in the case of prawns in nursery rearing and grow-out systems. In the present study marine yeast was incorporated in the diet of *P. monodon* which gave higher growth and survival than those fed on diets containing prawn meal or soybean meal. High amount of polyunsaturated fatty acids, high amount of vitamins and minerals present in marine yeast may be the reason for the better performance of *P. monodon* with this diet.

5.3.1.2 Food Conversion Ratio

In the present study, lower food conversion ratios were recorded in the case of prawns fed on diet containing animal protein sources than diets containing single cell or plant protein sources. The lowest food conversion ratio was recorded in the case of diet containing clam meal (2.18) followed by diet containing slaughter house waste meal (2.59). The highest food conversion ratio was recorded in the case of prawns fed on diet containing marine yeast (3.26).

Gopal (1986) reported that diet with plant protein sources gave relatively higher FCR values than those fed on diets with animal protein sources. Similar observation has also been recorded in the present study. Thus the diet containing the soybean meal recorded higher FCR values (3.12) than those containing animal protein sources.

Specific studies on feeds and nutrition of *P. monodon* conducted at Kakdwip fish farm by Rajyalakshi *et al.* (1979) have reported a food conversion ratio ranging from 2.4 to 6 for various feeds prepared from fish meal, shrimp meal and squid meal as main components. Mohammed Sultan *et al.* (1982) formulated feeds with frog flesh waste and reported a food conversion ratio between 3.01 and 4.96 for *P. indicus* and 5.87 and 8.21 for *P. monodon*. The food conversion ratio observed in the present study with clam meat was found to be superior than for those reported earlier.

Feeding trials conducted on *P. monodon* using a practical feed in the Philippines (SEAFDEC, 1981) have shown a food conversion ratio of 4.8. In a semi-intensive culture experiment with the same species (SEAFDEC, 1983), a commercial prawn feed with 45% protein and an experimental feed with 35% protein produced FCR of 3.4 to 4.6 and 6.1 respectively. In yet another intensive culture experiment (SEAFDEC, 1983), crustacean pelleted feed resulted in a FCR of 3.01 for P. monodon. In Philippines, Liu and Mancebo (1983) recorded a FCR of 1.69 to 1.78 in P. monodon using a commercial formula feed developed by the President Enterprises Corporation.

The higher FCR obtained for clam meal diet in the present study may be due to the nutritive quality of clam meal compared to the protein sources used in other diets. Ahamad Ali and Mohammed (1985) also reported that the diet prepared with clam meal gave the best FCR among the different diets used in his study.

5.3.1.3 Protein Efficiency Ratio (PER)

The present results indicate that prawns fed on pellets based on clam meal were more efficient in converting the dietary protein, followed by the pellet based on prawn meal. It was also found that feeds based on animal protein sources gave better PER values than those based on plant or single cell protein sources.

Steffens, (1981) reported that the PER values can be used to evaluate the quality of protein in the diet, those with high PER values, are of good quality protein and those with low PER values are of poor quality. Hence in the present study, it can be surminsed that the feed based on soybean meal which recorded low PER values is of lower protein quality, compared to other feeds tested.

5.3.1.4 Protein Digestability Coefficient

The digestability of different components of a feed depends on many factors. The difference in digestion coefficients have been attributed to size variations, microflora of intestine and digestive enzymes by many workers (Nail, 1962; Shell, 1967; Dabrowski, 1977). Digestability may be limited due to incomplete digestive action or because of lack of complete absorption (Maynard and Loosli, 1978).

The range in protein digestability in the present study was 84.39% and 92.22%, the coefficients being higher for pellets CM and SW, indicating better utilization of these diets by prawn. Nose and Toyama (1966) and Nomura _et al._ (1973) have reported that different proteins will have different digestability. Nose (1963) reports that an unfavourable amino acid composition of the diet may also affect the protein digestability.

5.3.2 Biochemical Composition of the Experimental Prawns

It is well known that chemical content of the diet influences the body composition of fish (Phillips _et al._, 1966; Jayaram and Shetty, 1980) and hence the study of biochemical composition of an organism is of paramount importance in formulation of its diet. Gopal (1986) reported that prawns show variations in body composition depending on the quality of protein in their diets. Variations in chemical composition of _P. monodon_ fed on diets based on different protein sources are discussed below.

Significant variations were observed for all the biochemical parameters studied except for the carbohydrate content in the experimental animals fed on diets based on different protein sources. The moisture content of the prawn muscle varied between 75.16% and 76.58%. The higher moisture content was observed in prawns fed on plant protein sources, than those fed on animal or single cell protein sources. Brandes and Dietrich (1958) and Love (1970) have pointed out that moisture shows an inverse relationship with fat content in fishes. A similar trend was observed in all the treatments of the present study also, except for prawn fed on diet containing marine yeast.

Data of protein content in the test animals indicate that prawns show preference to animal protein sources compared to plant or single cell protein sources for maximum protein synthesis. Among the prawns fed on diets with animal protein sources, clam meal (CM) based pellet, recorded the highest protein percentage (65.71%), which may be due to the quality of protein in the clam meat and its amino acid profile. According to Cowey and Forster, (1971) and Deshimaru and Shigueno (1972) growth and deposition of protein in the concerned organisms are based on the quality of protein intake. Gopal (1986) has reported higher growth rates and protein deposition in prawns fed on animal protein source because of their higher biological value over other protein sources. The lower protein synthesis in prawns fed on plant or single cell protein sources may be due to the absence of essential nutrients in these diets. Cowey and Corner (1963) reported that variation in essential as well as non-essential amino acids present in diet may affect the metabolic process which in turn influence the synthesis of body proteins.

Prawns fed on diet with animal protein sources had significantly higher fat content compared to those fed on diet containing single cell or plant protein sources. Maximum deposition of fat was observed in prawns fed on diet based on slaughter house waste, (16.9%) followed by those based on clam meal (16.10%). Venugopal (1980) reported that an increased dietary lipid level enhances the deposition of fat in fish flesh. A similar positive correlation between fat content of the diet and accumulated body fat was noticed in the present study too, where the fat content in the diet based on slaughter house waste meal was 9.72% followed by clam meal based diet (8.99%). Buckley and Groves (1979) also suggested that crude fat content in the feed stuff was the major factor which determines the fat content of the organism. The lower fat deposition in the prawns fed on diet based on plant proteins may be due to inhibitory effect of higher levels of fatty acids of W6 (linoleic) series as observed by Cowey and Sargent, (1972).

The ash content was relatively higher in prawns fed on diet based on plant or single cell protein sources than animal protein sources. The highest values was seen in prawns fed on pellets based on soybean meal (22.82%) followed by that on marine yeast (19.48%). Among the diets based on animal protein sources, prawn meal based diet gave the highest ash percentage (18.12%). The carbohydrate content of the prawns fed on different test diets ranged from 1.71 to 2.94%, with the maximum in diet based on soybean meal followed by that with marine yeast. This may be due to the higher amount of carbohydrate content in the respective diets. An inverse relationship between the protein and carbohydrate content of muscle as reported by Khawaja in 1966 is found to be true in the present study also.

Prawns fed with diet based on plant protein source had significantly lower calcium and phosphorus levels compared to those fed on diet based on animal or single cell protein sources. Among the prawns fed on diets based on animal or single cell protein sources, variation in phosphorus and calcium content is insignificant, indicating that utilization of these minerals is not affected by the dietary protein sources. The low phosphorus levels in the body of prawns fed on diet containing plant protein sources in comparison to the other two protein sources may be because of the fact that major part of the phosphorus in the plant product is found to phytic acid (Lall, 1978) which is usually unavailable to animals due to the absence of enzyme phytase (Muir and Roberts, 1982).

5.3.3 Oxygen Consumption, Ammonia Excretion and O : N Ratios

Oxygen consumption, ammonia excretion and O : N ratios can be used as an index of evaluation of a protein source, since these values vary with different protein sources tested. Increased oxygen consumption and reduced ammonia excretion rate at the same time with higher O : N ratios may indirectly indicate the efficiency of a diet. Laxminarayana and Kutty (1982) reported a reduced oxygen consumption and increased ammonia excretion rate for *P. semisulcatus* under stress conditions. Gopal (1986) is of the view that ammonia excretion rate is an indication of efficiency of protein metabolism in prawns. He has also reported that if a prawn is fed on good quality diet, the diet is efficiently utilized for better growth with minimum catabolism of protein, which in turn indicates the lower ammonia excretion rate. In the present study, lower ammonia excretion rate was observed in prawns fed on diet based on clam meal (0.0157 mg NH_3-N/g/l) and highest in prawns fed on diet containing soybean meal (0.0285 mg NH_3-N/g/l).

Capuzzo and Lancaster (1979) reported decreased O : N ratios in H. americanus which they attributed to the increased protein catabolism. Since increased protein catabolism has been reported in organisms fed on poor quality diet, the lower O : N ratios can be used as an index for diet of poor quality. In the present study the lowest O : N ratio was obtained in the prawns fed on diet containing soybean meal (5.27) and highest in those fed on diet based on clam meal (9.52). Ragnault (1981) observed a reduction in O : N ratios in Crangon crangon when it is subjected to continued exposure to stress conditions.

5.4 Study on Optimum Food Ration

In shrimp farming systems, determination of optimum food ration is of great significance in view of reducing the cost of operation, at the same time maintaining the environmental hygiene. Shellbourne et al. (1973) reported that in fishes both specific growth rate and food requirements expressed as percentage of body weight fed per day, declined as mean weight of the fish increased. If the ration levels expressed in this form are maintained constant above a minimum, they eventually become surplus for requirements of maximum growth.

Inorder to achieve an efficient feeding regime in an aquaculture system, continued adjustment of the ration level is necessary to compensate the changing requirement (Sedgwick, 1979). A precise knowledge of the relationship between food requirement and body weight for a particular species and diet would be essential to avoid both overfeeding and restricted growth through sub maximum rations.

In the present study, the optimum ration for prawns with an average size of 3.3 cm/0.209 g for the selected clam meal diet was found to be 6% of the body weight while maintenance ration and maximum ration were found to be 1.8% and 15% respectively. Venketaramiah et al. (1972) reported that the quantity of food offered to an animal ranges from 5-100% of the wet weight of the animal. Sick et al. (1973) observed that consumption rates in prawn were directly proportional to the animal size and length of exposure to any particular feed. Apud (1988) reported that the optimum ration for P. monodon having an average weight of 1.7 g is 6% of the body weight and maximum ration as 13% of the body weight. Sedgwick (1979) found that the optimum ration for P. merguiensis juveniles was 6% of the body weight while the maintenance and maximum rations were 2% and 16% of the body weight respectively.

5.5 Study on Optimum Feeding Frequency

In intensive farming operations feeding frequency has an important role in improving overall yeild and reducing the cost of production. A decline in feed efficiency may be incurred, if the food is left exposed in water for long periods before ingestion due to deterioration of the nutritive value through leaching of soluble components from the feed (Forster, 1972; Sedgwick, 1979). This problem can be resolved to some extent by presenting the diet to the prawns in a way which ensures rapid consumption which is mainly achieved through frequent feeding practice. Feeding the animals in divided doses twice or thrice daily was found to contribute towards better growth and conversion efficiency in prawns than feeding the entire dose once a day.

In the present study food consumption, growth and survival of P. monodon juveniles increased as the feeding frequency was raised from once a day to thrice daily, but beyond this level these parameters showed a declining trend. Almost similar observations were reported by Marian et al. (1987) in Macrobrachium lamerei. Sampath and Srithar (1987) reports that in P. monodon, rate of feeding, absorption, and production were increased as the frequency of feeding was raised from once in five days to twice a day. A further increase in feeding frequency however did not influence these parameters. However, Sedgwick in 1979 reported that better growth and feed efficiency can be achieved by increasing the feeding frequency from once a day to four times daily in P. mergiuensis. Apud et al. (1980) reports that better growth and survival in P. monodon can be achieved by manipulating the feeding frequencies, i.e. twice daily, during the first month, three to four times daily in the second month and five times daily during the third and fourth months.

Primavera et al. (1979) reports that food consumption rate increases if the food is given during afternoon and evening hours compared to morning hours, with three hours feeding frequency showing a major peak at 1800 hours (dusk) and minor peak at 1200 hours (noon). Similar observation was also noticed in the present study, where food consumption and growth were higher when prawns were fed during night hours than during day time. Pascual (1986) also reports that best feeding time is late afternoon and evening hours for P. monodon since prawns move around the perimeter of the pond during this time of the day.

VI. SUMMARY

VI SUMMARY

The present investigation has been carried out to evaluate the efficiency and keeping quality of pelleted feeds formulated from different protein sources viz. clam meal, prawn meal, slaughter house waste meal (animal proteins), soybean meal (plant protein) and marine yeast (single cell protein), for rearing the juveniles of Penaeus monodon. The main observations were as follows:

1 Proximate analysis of the various ingredients used in the study, revealed that clam meal and slaughter house waste meal contained more crude protein than soybean or marine yeast.

2 The protein levels in the formulated diets were almost equal (39.76% to 40.30%), crude fat was found to be more in pellet SW (9.72%) than the rest of pellets. Maximum carbohydrate content was recorded in pellet SM (29.38%) and least in pellet PM (19.80%). Crude fibre content was high in pellet SM (7 .02%). All the diets were isocaloric, the caloric content ranged from 3.34 K.cal/g (pellet MY) to 3.72 K.cal/g (pellet CM).

3 The quality of formulated diets over a period of four months of storage remained good with a slight decrease in nutritive value.

4 The pellet SM was found to be more water stable at the end of three hours,followed by the pellet CM and SW.

5 The variations observed on the water quality parameters were found to be well within the tolerance limits indicating that supplementary feeding had no adverse effect on the quality of water if properly maintained.

6 At the end of 42 days of rearing, the highest growth rate and survival were recorded in P. monodon fed on diet based on animal protein source than diet based on plant protein source. Among the animal protein sources tested, the diet based on clam meal proved to be the best followed by diet based on slaughter house waste. Diet based on marine yeast recorded superior growth performance than those based on prawn meal or soybean meal.

7 Diets based on animal protein sources were found to give better food conversion ratios than those based on plant or single cell protein sources. The lowest food conversion ratio (2.18) was recorded in prawns fed on diet containing clam meal (CM) followed by diet containing slaughter house waste meal (SW). The highest FCR (3.26) was recorded in prawns fed on diet based on marine yeast (MY).

8 Protein efficiency ratio and protein digestability values were found to be higher in prawns fed on diets based on animal protein sources than those based on plant or single cell protein sources. Among the diets tested, the diet based on clam meal recorded highest PER (1.13) and protein digestability (92.22).

9 The biochemical composition of prawns reared on different feeds show significant ($P < 0.05$) variations in moisture, protein, fat and ash content but no significant difference was noticed in the case of carbohydrate content of the prawns.

10 Oxygen consumption, ammonia excretion O : N ratios were significantly ($P < 0.01$) influenced by the protein sources. Oxygen consumption rate

of prawns fed on diet with animal protein sources was found to be higher than those with single cell protein source. Lower ammonia excretion rate was recorded in prawns fed on diets based on animal protein sources than those based on plant or single cell protein source. The highest O : N ratio was recorded in prawns fed on diet based on animal protein source than plant or single cell protein sources. Among the different diets tested, the diet based on clam meal recorded the highest oxygen consumption rate (o.1705 mg O_2/g/hr) lowest ammonia excretion (0.0157 mg NH_3-N/g/hr) and highest O : N ratio (9.52).

11 The optimum food ration for *P. monodon* juveniles was found to be at 6% of the body weight while the maintenance and maximum rations were 1.8% and 15% respctively for clam meal based diet.

12 Food consumption, growth and survival of *P. monodon* increased as feeding frequency was raised, however these parameters increased only upto an increase of 3 meal/day and there after a declining trend was observed.

VII. REFERENCES

VII REFERENCES

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GROWTH RESPONSES OF *PENAEUS MONODON* FABRICIUS TO PELLETED FEEDS OF DIFFERENT PROTEIN SOURCES

BY

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ABSTRACT

The efficiency and keeping quality of the five pelleted feeds formulated from different protein sources viz; clam meal, prawn meal, slaughter house waste meal, soybean meal, single cell protein (marine yeast) were tested with a view to develop a suitable supplementary feed for *Penaeus monodon.*

The feeds were isonitrogenous, crude protein content ranged from 39.76 to 40.3%) and isocaloric (Caloric value between 3.34 - 3.72 K.cal/g). The quality of the feeds over a period of four months of storage remained good, eventhough a slight reduction in nutritive value has occurred.

P. monodon juveniles were reared for 6 weeks in experimental tanks with different artificial feeds, the highest growth and survival were recorded in those fed with diet based on animal protein sources. Among the feeds tested, the clam meal based diet was found to be superior followed by diet based on slaughter house waste meal. The diet based on marine yeast was found to give better performance than those based on prawn meal or soybean meal.

The food conversion ratios obtained with different feeds ranged from 2.18 to 3.26. The lowest FCR was recorded in diet based on clam meal (2.18) and the highest (3.26) in diet based on marine yeast.

Protein efficiency ratios and protein digestability values were found to be higher in prawns fed with diet based on clam meal followed by diet based on slaughter house waste meal.

Significant variations in the biochemical composition of prawns fed with diet based on different protein sources were recorded.

Significant variations in oxygen consumption, ammonia excretion and O : N ratios were recorded in prawns reared on different protein based diets. Highest oxygen consumption rate and lowest ammonia excretion rate were observed for diets based on animal protein sources than plant or single cell protein source. Among the different diets tested, diet based on clam meal recorded highest oxygen consumption, lowest ammonia excretion and highest O : N ratio values.

The optimum food ration for P. monodon juveniles was found to be at 6% of the body weight while maintenance and maximum rations were 1.8% and 15% respectively for clam meal based diet.

Food consumption, growth and survival of P. monodon increased, as feeding frequency was raised, however these parameters increased only upto an increase of 3 meal/day and there after they showed a declining trend.