EVALUATION OF SUPPLEMENTARY FEEDS AND OPTIMUM RATION FOR *CHANOS CHANOS* (FORSKAL) FRY

By

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THESIS

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DEDICATED TO

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I hereby declare that this thesis entitled "EVALUATION OF SUPPLEMENTARY FEEDS AND OPTIMUM FOOD RATION FOR *Chanos chanos* (FORSKAL) FRY" 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
1	INTRODUCTION	1
2	REVIEW OF LITERATURE	4
	2.1 Nutritional Requirements of Warmwater Finfishes with special reference to Milkfish	4
	2.1.1 Proteins.	5
	2.1.1.1 Essential Amino Acids.	6
	2.1.2 Lipids.	7
	2.1.2.1 Essential Fatty Acids.	9
	2.1.2.2 Phospholipids.	10
	2.1.3 Carbohydrates.	10
	2.1.4 Vitamins.	11
	2.1.5 Minerals.	12
	2.2 Food and Feeding habits of *C. chanos*	13
	2.3 Supplementary Feeding in Brackishwater Finfishes	15
	2.4 Protein Sources used in Supplementary Feeding	19
	2.4.1 Protein sources of Animal origin.	19
	2.4.2 Protein sources of Plant origin.	22
	2.4.3 Mixed protein sources.	25
	2.4.4 Unconventional Protein sources.	27
	2.5 Studies on O N Ratio	29
	2.6 Studies on Optimum Food Ration	30
3	MATERIALS AND METHODS	33
	3.1 Preparation of Feeds	33
	3.1.1 Feed Ingredients.	33

3.1.2 Proximate composition of Feed Ingredients 34

3.1.3 Formulation and Preparation of test diets 35

3.1.4 Proximate composition of the Formulated Feeds 37

3.2 Sinking rate of Feed Crumbles 37

3.3 Water Stability of Formulated Feeds 37

3.4 Microbiological quality of Formulated Feeds 38

3.5 Keeping quality of Formulated Feeds 38

3.6 Experimental Milkfish fry and their acclimatization 38

3.7 Experimental tanks and water quality management 40

3.8 Study to Evaluate the Supplementary Feeds 40

3.8.1 Determination of Digestibility co-efficient 42

3.8.2 Monitoring of Physico-Chemical parameters. 42

3.8.3 Recording of Observations. 43

3.8.3.1 Food Conversion Ratio. 43

3.8.3 2 Protein Efficiency Ratio 43

3.8.3.3 Protein and Lipid Digestibility co-efficients. 43

3.8.3.4 Survival rate. 44

3.8.3.5 Growth. 44

3 8 3.6 Specific Growth Rate. 44

3.8.4 Biochemical analysis of Carcass . 45

3.8.5 Estimation of O N Ratio 45

3.9 Study on the Optimum Food Ration 46

3.10 Statistical Analysis 46

4 RESULTS 47

4.1 Proximate composition of Feed Ingredients and Formulated Feeds 47

4.1.1 Feed Ingredients 47

4.1.2 Formulated Feeds 47

4 2 Sinking rate of Formulated Feeds 52

4.3 Water Stability 52

4 4 Microbiological quality of Formulated Feeds 54

4 5 Evaluation of Supplementary Feeds 54

4 5 1 Water quality maintenance. 54

4.5.1.1 Temperature. 54

4.5.1.2 p^H 56

4.5.1 3 Dissolved Oxygen. 56

4.5.2 Efficiency of Feeds. 56

4.5.2.1 Food Conversion Ratio. 56

4.5 2.2 Protein Efficiency Ratio. 61

4.5.2.3 Protein Digestibility co-efficient. 61

4 5 2 4 Lipid Digestibility co-efficient 61

4.5.3 Biochemical composition of C chanos fry. 67

4 5.4 Oxygen consumption, Ammonia excretion and O N Ratio. 70

4.5.5 Performance of various feeds based on Survival and Growth of C chanos fry 76

4.5 5.1 Based on Rate of Survival. 76

4 5 5.2 Based on Growth. 76

4 5.5 3 Based on Specific Growth Rate 81

4.6 Studies on Optimum Food Ration 85

4.6 1 Water quality. 85

4.6.2 Rate of survival 85

4 6.3 Specific Growth rate 85

4.6.4 Food Conversion Ratio 88

4.6.5 Extrapolation of Optimum Food Ration from graph. 89

5 DISCUSSION 92

5.1 Keeping quality of Formulated Feeds 92

5.1.1 Moisture. 92

5.1.2 Protein. 93

5.1.3 Fat. 94

5.1.4 Ash, Crude fibre, Carbohydrate. 94

5.1.5 Total Heterotrophic Bacteria and Fungi. 95

5.1.6 Water Stability. 96

5.2 Water quality parameters in relation to Growth and Supplementary Feeding 97

5.3 Evaluation of Supplementary Feeds 97

5.3.1 Based on Efficiency of Feeds. 97

5.3.1.1 Food Conversion Ratio. 97

5.3.1.2 Protein Efficiency Ratio. 99

5.3.1.3 Digestibility co-efficients. 99

5.3.2 Based on Biochemical composition of C. Chanos fry before and after Feeding. 104

5.3.3. Based on Oxygen consumption, Ammonia excretion and O N Ratio. 106

5.3.4 Based on Growth Studies. 108

5.4 Optimum Food Ration 113

6 SUMMARY 115

7 REFERENCES 118

8 ABSTRACT

LIST OF TABLES

		Page
1.	Percentage composition of different ingredients and their protein contribution in formulated feeds.	36
2.	Proximate composition of the ingredients used in the feed formulation.	48
3.	Proximate composition of formulated feeds.	50
4.	Effect of storage on the keeping quality of formulated feeds.	51
5.	Range of temperature (in °C) of the experimental tanks during the study for the evaluation of formulated feeds.	55
6.	Fluctuation of pH in the experimental tanks during the study for the evaluation of formulated feeds.	57
7.	Variations in dissolved oxygen content in the experimental tanks during the study for evaluation of formulated feeds.	58
8.	Food conversion ratio of C.chanos fry fed with various formulated feeds.	59
9.	Protein efficiency ratio of C.chanos fry fed with various formulated feeds.	62
10.	Protein digestibility co-efficient of various formulated diets fed to C.chanos fry.	64
11.	Lipid digestibility co-efficient of various formulated diets fed to C.chanos fry.	66
12.	Biochemical composition of C.chanos fry fed with various formulate feeds.	68
13.	Oxygen consumption, ammonia excretion and O N ratio in C.chanos fry fed on formulated feeds compounded from various protein sources.	71
14.	ANOVA Table for efficiency of formulated feeds fed to C chanos fry.	72
15.	Rate of survival of C.chanos fry fed on different formulated diets.	77

16. Average length (cm) and weight (g) gain, and percentage increase in growth attained by C.chanos fry fed with various diets for 42 days. 79

17. Specific growth rate of C.chanos fry fed with various diets for 42 days. 82

18. ANOVA Table for the performance of feed on survival and growth of C.chanos fry. 83

19. Water quality parameters of experimental tanks during the study for optimum food ration in C.chanos fry. 86

20. Effect of food ration on survival, growth and food conversion ratio in C.chanos fry. 87

21. ANOVA Table - optimum food ration for C.chanos fry. 89

LIST OF FIGURES

Page

1. The effect of storage on the proximate composition of feeds. 53

2. Food conversion ratio and specific growth rate of C.chanos fry fed with different formulated feeds for 42 days. 60

3. Protein efficiency ratio of various supplementary feeds fed to C.chanos fry. 63

4. Protein and lipid digestibility of various formulated feeds fed to C.chanos fry. 65

5. Comparison of biochemical composition of C.chanos fed on various formulated feeds. 69

6. Gain in length and weight of C.chanos fry fed with various formulated feeds. 80

7. Graph showing the specific growth rate and food ration of C.chanos fry fed on clam meal diet for 21 days. 91

LIST OF PLATES

		Page
1.	Average initial size of *Chanos chanos* fry used for the evaluation of formulated feeds.	39
2.	Experimental set up	41
3.	Comparison of growth attained by *C.chanos* fry fed on various formulated feeds.	78
4.	Schooling behaviour of *C.chanos* fry during feeding.	103

INTRODUCTION

1 INTRODUCTION

The last decade has witnessed the art of pisciculture being oriented in scientific lines in different parts of the world. The overdependence on shellfishes for aquaculture is fast relegating fish culture to a lower slot, especially, in maritime nations. Nevertheless, finfishes still contribute to a major share (49.22%) of the world aquaculture production of 14.47 million tons (FAO, cited by Conrads, 1991). If the present trend continues, milkfish alone will contribute to a major share, since at present it forms 3.1% (Nash, 1988) of the total world aquaculture production.

Milkfish, Chanos chanos, the only member of family Chanidae, is the most widely used warmwater finfish for culture in the traditional brackishwater aquaculture systems throughout Asia and Far East. It is a fast swimmer, herbivore, and can tolerate wide ranges of salinity (0-158 ppt), temperature (8.5 - 42.7°C), and low dissolved oxygen contents (Chen, 1990). Its fast growth, high resistance to diseases and easy adaptation to supplementary feeding, make it one of the best candidate species for aquaculture. At present, traditional milkfish culture is gradually shifting to semi-intensive or more intensive culture systems (Sumagaysay and Chiu, 1988, Chen, 1990).

However, commercial hatchery production of the seed of milkfish has not been perfected and its farming is mostly dependent upon natural seed. The seasonal availability of its seed in nature necessitates an efficient nursery rearing practice for making available the maximum number of quality seed for culture purpose It has also made essential, formulation of fry feeds, which gives good survival, growth and food conversion rates, either to be used as supplementary feeds in ponds or as complete diet in intensive systems

The productivity of any fish culture system and hence its profitability depends to a great extent on the amount of supplementary feeds used. The more intense the system, the greater is the importance of supplementary feed and higher is the proportion of feed cost to the total production cost (Hepher, 1988). The cost of feed comes to about 40 - 60% of the total production cost (FAO, 1983) Most of the fish feeds available in Asia are formulated from imported items like fish meal, corn, and wheat middlings and so their prices are high Hence the recent trend is to minimize feed cost by substituting or complementing fish meal with locally available feedstuffs or with unconventional protein sources It has created the problem of feed acceptability and palatability (Tacon and Jackson, 1985), especially by the younger stages of fish. Cost savings at the expense of nutritional quality in fry feed is a false economy, since fry stage is the one where greater relative gain in growth can be made by feeding high quality feed, which will persist upto harvest, with least total investment (Hardy, 1989) Tacon and Jackson (1985) are also of the opinion that the development of high quality protein sources in addition to fish meal will reduce the reliance of fish meal in fish feed manufacturing industry and ensure the fish farmers a relative stable and high quality ration. With this in view, certain feeds compounded from high quality protein sources like fish meal, squid meal, clam meal, prawnhead waste and soyabean meal were tested in C chanos fry Once these feeds are found suitable, they can be put to use in the nursery rearing of milkfish fry, according to the market price, local availability, nutrient composition of each diet or on the intensity of culture The effect of storage on the keeping quality of the formulated feeds were also tested.

The objective of fish feed formulation is to supply the required nutrients for optimum animal production (Conklin et al , 1977) Once a nutritionally adequate feed is developed, optimum food production can be obtained by regulating the feeding level Feeding fish too little will result in the utilization of most of the feed for maintenance Conversely, feeding too much will result in low utilization and wastage of food. In both these cases food conversion rate will increase and the profitability will decrease. Hence an attempt has also been made in the present study, to find out the optimum food ration of C chanos fry with the selected feed of the first experiment.

REVIEW OF LITERATURE

2 REVIEW OF LITERATURE

2.1 Nutritional Requirements of Warmwater Finfishes with special reference to Milkfish

Eventhough artificial feeding of warmwater fishes has become popular in recent years, studies on their nutritional requirements are much limited than those of coldwater fishes Most of the purified diets which have been developed for warmwater fishes are modified formulations of those diets which are successfully used for trout and salmons (Dupree and Sneed, 1966).

The first purified diet used in the nutritional studies of C.chanos was developed by Lee and Liao (1976). They recommended vitamin-free casein (about 60% of the diet) supplemented with 0 5% L-tryptophan as a better protein source for young milkfish than a combination of casein and gelatin, soyabean oil (10% of the diet) was a better lipid source than cod liver oil, vitamin and mineral mixtures at 4% and 10% of the diet, respectively, recommended for chinook salmon (Halver, 1957) were also found satisfactory for milkfish. This purified diet also contained dextrin as carbohydrate source and a high level of carboxymethyl cellulose (10%) as a binder However, Camacho and Bien (1983) reported that a purified diet with 40-50% vitamin-free casein and 12-15% gelatin to be the best source of protein for milkfish fry For the normal growth of fry, 8-10% fat, and 3-4% vitamin mixture must be added to this purified diet. Recently, Teshima et al (1984) also developed purified diets for milkfish fingerlings. They found that high growth occurred when a diet containing 35% casein and 15% gelatin supplemented with 0.5% methionine and 0.5% tryptophan was fed at 30 to 50% of fish biomass daily

2.1.1 Proteins.

Protein is the basic component of animal tissues and is therefore an essential nutrient for both maintenance and growth The capacity of fish to synthesise amino acids denovo from carbon skeltons is limited and most of the proteins must therefore be supplied through the diet (Hepher, 1988).

The optimum dietary level of protein required for maximum growth of farmed fishes is 50-300% higher than that of terrestrial animals (Cowey, 1975). In a study by Lim et al. (1979), it was found that wild milkfish fry with 40 mg body weight required 40% dietary protein for maximum growth, efficient conversion and high survival, when fed at 10% of fish biomass daily, at a salinity of 32-34 ppt and temperature 25-28°C. The test diet contained casein as protein source, dextrin as carbohydrate source, equal parts of cod liver oil and corn oil as lipid source, and vitamin and mineral premixes. The protein requirement of milkfish is seen to be near the values reported for other warmwater fishes particularly *Oreochromis mossambicus* (40% - Jauncey , 1982), *Liza parsia* (40% - Kiran, 1989), *Cyprinus carpio* (38% - Ogino and Saito, 1970) and *Ctenopharyngodon idella* (41-43% - Dabrowski, 1977). In other fishes it shows a range of 35-55% of the diet (Easterson, 1987 Pandian, 1987, 1989, Hepher, 1988, Wilson, 1989). This wide variation may be because, even within the same species protein requirement varied with the size of fish, water temperature and salinity, stocking rate, feeding regime and culture practices (Hepher, 1988). The optimal dietary protein level for fish is also influenced by an optimal dietary protein to energy balance, the amino acid composition and digestibility of test proteins, and the amount of non-protein energy sources in the test diet (Wilson, 1989).

The influence of changes in dietary protein to energy ratios on growth and protein utilization has been demonstrated in several species of fish, viz., yellow tail (*Seriola quinqueradiata*) (Takeda *et al*., 1975) estuarine grouper (*Epinephelus salmoides*) (Teng *et al* , 1978), common carp (*Cyprinus carpio*) (Takeuchi *et al*., 1979) *Oreochromis aureus* (Winfree and Stickney, 1981), and *O.mossambicus* (Jauncey, 1982). A study on the protein - energy requirement of *C.chanos* showed that 30-40% of dietary protein, 10% lipid, and 25% carbohydrates were required by milkfish fingerlings, weighing 0 5 to 0.8 g (Pascual, 1984) Hepher (1988) has calculated the protein energy ratio of milkfish fry as 110 mg protein/Kcal of energy based on the experiment done by Lim *et al* (1979).

2.1.1.1 Essential Amino Acids.

Fish, like other animals do not have a true protein requirement but has a requirement for a well balanced mixture of essential and non-essential amino acids Essential Amino Acid (EAA) requirement by fish has been reviewed by Wilson (1985, 1989) The complete amino acid requirement for Japanese eel and channel catfish has been established. A limited number of requirement values have also been reported for gilthead bream, sea bass, and Oreochromis mossambicus (Wilson, 1985) The fish species so far studied, have been shown to require the following amino acids, viz., arginine, histidine, isoleucin, lysine, methionine, phenylalanine, threonine, tryptophan and valine (Wilson, 1985, 1989, Halver, 1989) The EAA requirement of fishes have been shown to correlate well with EAA pattern of their own whole body tissue (Wilson and Poe, 1985).

In milkfish, Coloso et al. (1983) have calculated a reference amino acid pattern by determining the amino acid composition of precipitable proteins from the whole body of milkfish fry (7 1 mg initial weight) reared on Artemia salina nauplii and Brachionus sp , at weekly intervals. Their studies have shown lysine, leucine and arginine may be the first limiting amino acids for milkfish. This reference pattern could form an indirect basis for estimating the EAA of milkfish, once one essential amino acid is known (Cowey and Tacon, 1981, Santiago, 1985).

2.1.2 Lipids

The two main functions of lipid in fish body are energy provision and biomembrane stability. Fishes rely on lipids than on carbohydrates for energy purposes The neutral lipid component of fish is therefore an useful entity in diet preparation and is particularly desirable in feeds of fry/fingerlings, which require high energy intake for rapid growth. A number of reviews on the requirement of lipid in fishes are available (Cowey and Sargent, 1972, 1977, 1979, Hashimoto, 1975, Castell , 1979, Watanabe, 1982, Millikin, 1982, Kanazawa, 1985).

Warmwater fishes have improved lipid digestibility, absorption and utilization than coldwater fishes. The more omnivorous carp can utilize lipid efficiently as a dietary energy source (Takeuchi et al , 1979). Carnivorous fishes can also utilize lipid in their diet, provided adequate amounts of choline, methionine, and tocopherol are provided in their ration.

The dietary requirement of lipid for C.chanos fry is 8-10% from corn oil and cod liver oil in 1 1 ratio (Camacho and Bien, 1983) and 7-10% for fingerlings (0.83 g mean body weight) from cod liver oil alone (Alava and de la Cruz, 1983) Similar values have been reported for yellow tail (Seriola quinqueradiata) (Deshimaru et al , 1982) and Liza parsia (Kiron, 1989) However, Paulraj and Thirunavakkarasu (1987) have reported 6% lipid level to be optimum for the best utilization of food, protein and for growth in milkfish fry.

The sparing effect of dietary protein by lipid has been examined in yellow tail (Takeda et al , 1975), common carp (Sin, 1973 Viola and Rappoport, 1979), Oreochromis aureus (Winfree and Stickney, 1981), and striped bass (Millikin, 1983)

2.1.2.1 Essential Fatty Acids

Numerous studies have established the requirement of n-3 Poly Unsaturated Fatty Acids (PUFA) for fish in general, but the extent to which n-6 series of PUFA are essential, remains to be established However, n-6 PUFA may be required in small amounts especially for marine fishes (Sargent et al., 1989). The importance of Essential Fatty Acids (EFA) in fish nutrition has been emphasised by Sinnhuber (1969), Cowey and Sargent (1979) Watanabe (1982) Kanazawa (1985), Bell et al. (1986) and Sargent et al (1989).

Since fish are incapable of de novo synthesis of 18 2n-6, 18 3n-3, 20 5n-3, and 22 6n-3 acids, dietary sources of these fatty acids are likely to be essential for normal growth and survival. Unlike freshwater fishes, marine fishes lack the ability to chain elongate and desaturate n-3 PUFA and hence require higher n-3 PUFA, especially 20 5n-3, in their diet (Kanazawa, 1985, New, 1987) Experiments of Benitez and Gorriceta (1983) have indicated different fatty acid composition of milkfish body tissue reared in high saline and low saline ponds, indicating that milkfish may require varying amounts of EFA in their diets depending on the salinity

The experiments of Bautista and de la Cruz (1988) using purified test diets have indicated that in milkfish (1.6 g body weight), both linolenic (18 3n-3) and linoleic (18 2n-6) acids are effective for promoting high growth and survival rates. Although highest growth was attained by fish fed 1% linolenic acid it was not significantly different from fish fed with a combination of linolenic and linoleic acids. They also have shown that cod liver oil (high in linolenic acid) and corn oil (high in linoleic acid) are good sources of lipids for milkfish fry and fingerlings. Cod liver oil and coconut oil also promoted higher growth rate in milkfish fingerlings than beef tallow and coconut oil or pork lard and coconut oil (Alava, 1986). Although there are indications that milkfish have the ability to use linolenic or linoleic acids as precursors for the biosynthesis of long-chain PUFA of n-6 and n-3 series, (Gorriceta, 1982, Villegas et al., 1983) it remains to be shown whether long-chain PUFA have better growth enhancing effects than with linoleic and/or linolenic acids.

2.1.2.2 Phospholipids.

Kanazawa et al (1983), report that the dietary sources of phospholipids are essential for normal growth and survival of fish larvae of ayu and red sea bream. Among phospholipids lecithin alone or lecithin in combination with phosphatidyl inositol produce better growth rate and survival in these fishes.

2.1.3 Carbohydrates.

The carbohydrates are usually the cheapest source of energy (New, 1987). However, in fish carbohyrates are inferior to lipids in energy provision (Nagai and Ikeda, 1972) The omnivorous and herbivorous fishes adapt to utilization of high carbohydrate diets (Shimeno et al , 1981), while in carnivorous fishes its digestibility is low (Nagai and Ikeda, 1972). Carbohydrate utilization in fishes is also temperature dependent (Shcherbina and Kazlaukene, 1971).

According to Furukawa and Ogasawara (1952), a 5% cellulose addition in fish diets has a favourable effect on the nitrogen retension and body growth in common carp. The sparing effect of carbohydrates over proteins has also been reported in red sea bream (*Chrysophrys major*) (Furuichi and Yone, 1971), and common carp (Ogino *et al.*, 1976)

Reports on the carbohydrate requirement and utilization in *C.chanos* are scarce, except for the observation of Pascual (1984) that fingerlings of milkfish (0.5 - 0 8 g) require 25% carbohydrate, 10% lipid and 40% protein in their diet for best growth, and survival The ability of *C chanos* to digest cellulose is suspected, as Chiu and Bentitez (1981) could not find any cellulase activity in the intestine of *C chanos* Sumagaysay (1988) has reported that fibre has some energy value in *C.chanos*.

2.1.4 Vitamins.

The requirement of vitamins for warmwater fishes has not been investigated extensively. In general fishes require four fat-soluble vitamins (A, D. E and K) and eleven water-soluble vitamins. Of these, thiamine, riboflavin, pyridoxine, pantothenic acid, niacin, folic acid and vitamin B_{12} are required in small quantities, and function as co-enzymes, while myo-inositol, choline and biotin are required in higher quantities.

Vitamin requirement of fish vary with species, age, size and growth rate, physiological conditions especially at times of wound healing and stress. Some fishes have the ability to synthesise vitamins from glucose substrates or amino acids, while in some others intestinal microflora can synthesise vitamins (Halver, 1979, Hepher 1988). These sources reduce the dependence of dietary sources for vitamins in fishes.

2.1.5 Minerals.

The dietary mineral requirement of fish has been reviewed by Cowey and Sargent (1972), Nose and Arai (1976) and Lall (1989). In general, minerals required by fish are Calcium, Magnesium, Phosphorus, and a number of trace elements like Iron, Copper, Iodine, Manganese, Selenium, Zinc, Chromium, Cobalt, Boron and Molybdenum.

The Calcium requirement of fish is affected by water chemistry, Phosphorus levels in the diet, and species differences. Requirement of the Phosphorus is the highest (0.5 - 0.9% available phosphorus) of all minerals. However its level is not affected by dietary Calcium levels (Watanabe et al., 1988, Lall, 1989).

Usually, Calcium, Magnesium and Zinc that are required by fish can be provided by the feed ingredients and there is no need for the supplementation of these minerals. Selenium can be absorbed from water. The dietary requirement values of Iodine and Chromium have not been established in fishes (Watanabe et al., 1988). The dietary requirement of Iron vary from 150-170 mg, Mn 12-13 mg, Co 5-10 mg and Cu 3-4 mg/Kg of diet Boron and Molybdenum have been shown to improve the growth in carp (George, 1970).

Very little is known about the vitamin and mineral requirements of milkfish. Nevertheless, various vitamin and mineral premixes, such as those intended for the coldwater fish, chinook salmon (Halver, 1957) and for other warmwater fishes (NRC, 1977) have been used for milkfish (Lee and Liao, 1976, Lim et al., 1979, Santiago et al., 1983, Pascual, 1983) with satisfactory results.

2.2 Food and Feeding habits of C.chanos

The main food ingested by adult milkfish in the wild consists of benthic and planktonic organisms like gastropods, lamellibranchs, foraminiferans, filamentous algae, diatoms, copepods and nematodes (Hiatt, 1944, Tampi, 1958, Schuster, 1960, Alamanzan, 1970, Esguerra, 1975, Poernomo, 1976, Villaluz et al., 1976, Vincencio, 1977)

Food ingested by milkfish juveniles vary with habitat Wild milkfish fry and fingerlings feed on benthic, epiphvtic and planktonic organisms, while the most favourable food of milkfish fry and fingerlings under captivity is lab-lab ie., the biological complex composed of microbenthic plants and animals closely associated with the mud of pond floor like, various forms of bacteria, unicellular and filamentous blue-green and green algae, diatoms, protozoans, copepods, ostracods, some free-living flat and round worms, molluscs in their larval stages and some crustaceans. Filamentous green algae, as well as other higher aquatic plants, become suitable food for the milkfish only when they are in a partially decayed stage because they are soft and can easily be macerated (Schuster, 1960) The fish probably may, also benefit from the microorganisms growing on decayed material.

Benthic diatoms and blue-green algae were found to be the most desirable food for all age groups of milkfish in brackishwater ponds (Tang and Huang, 1966). Pantastico et al. (1986) found Oscillatoria alone or in combination with Chrococcus to be better food for C.chanos than Navicula alone.

Detritus also accounts for a large portion of the gut contents of adult fish (Tampi, 1958, Poernomo, 1976, Villaluz et al., 1976). Although detritus is the major food ingested by wild milkfish fry (Banno, 1980) and juveniles (Buri, 1980, Kumagai and Bagarinao, 1981, Trino and Fortes, 1989), the type and quantity of detritus may have an influence on its acceptability and value as food for milkfish (Santiago, 1986)

Thus milkfish can be considered planktonic, epiphytic, benthic or microphagous herbivorous feeder. According to Schuster (1960), this is because milkfish can resort to facultative feeding depending on the availability of food.

The feeding rhythm of milkfish has been studied by Schuster (1960), Banno (1980) and Chiu et al (1986 b). Milkfish in all stages seek food at day time (0600 to 1900 hrs) with peak feeding at 0700 to

1300 hrs (Banno, 1980) and the gut is completely devoid of food between 2200 and 0200 hrs (Chiu et al., 1986 b). This is in accordance with the findings of Chiu and Benitez (1981) that intestinal amylase activity peaked at about noon (1230 hrs) when gut was full and was lowest at 0030 hrs when gut was empty. However Chiu et al (1986 b) have shown, that providing supplementary feeds in the early morning and late afternoon can result in a shift of the feeding pattern from a sharp noon peak to a lower peak of longer duration in milkfish, provided dissolved oxygen is adequate.

2.3 Supplementary Feeding in Brackishwater Finfishes

In recent years, great importance is being paid to scientific culture of brackishwater finfishes, especially in South East Asia, in order to improve the protein sources as well as employment opportunities in the rural areas. Regular supply of nursery reared healthy fingerlings is the most essential pre-requisite for any fish culture system Since stocking densities are higher in scientific culture systems, natural food must be supplemented with artificial diets for obtaining good growth rate in fishes. However, supplementary feed is not extensively used in brackishwater culture systems as in freshwater pisciculture (Karmakar and Ghosh, 1984) Inspite of this, several empirical feed formulations are in use, which are improved versions of grow-out diets for same species or related species (Paulraj, 1989).

Supplementary feeds used in various countries have been mentioned by Ling (1967) - in Honkkong, Lin (1969) and Chuang *et al.* (1986) - in Taiwan, Noor-Hamid and Mardjono (1976), and Noor-Hamid *et al.* (1977) - in Indonesia, Sarig (1981) - in Israel, Villaluz *et al.* (1982) - in Philippines, Pathmasothy (1983) - in Malaysia, and Paulraj (1987) - in India.

The early formulations of feed for brackishwater finfishes were based on attempts to duplicate the composition of natural foods, this included the use of live-food organisms like phytoplankters and zooplankters for rearing the fry and fingerlings of milkfish and mullets (Alikunhi *et al.*, 1975, Ranoemiharjdo *et al.*, 1975, Vincencio, 1977, Acosta and Juario, 1983, Pantastico *et al.*, 1986), and tilapia (Pantastico *et al.*, 1985). Various unicellular algae and yeast Single Cell Proteins (SCP) are also used as live-food organisms for a variety of fishes (Aplebaum, 1979, Watanabe *et al.*, 1983). 'Digman' a collection of hydrophilic plants like *Ruppia*, *Najas*, *Halophilla* and *Thalassia*, and *Eichhornia crassipes* in fresh or dired form, and *Gracillaria* - a marine red algae-formed the food of *C. chanos* in Philippines (Bardach *et al.*, 1972). Aquatic weeds like *Enteromorpha* and *Hydrilla* were found to be suitable feed for *Etroplus suratensis* (Sathiavathy, 1989). Chopped grasses, water plants, algae, papaw and banana leaves formed the feed of tilapia (Huet, 1975, Maar *et al.*, 1966, Ling, 1967). Apart from these natural diets, various other fresh - wet diets were also used especially for the culture of carnivorous fishes, they included offal from slaughter houses, fish markets and fish processing plants, beef blood, earthworm, and silkworm pupae for eel

(Ling, 1967) liver, blood and glandular tissue of animals for silversides (Odontesthes sp) (Ghittino, 1972), whole or chopped trash fish for sea bass (Lates calcarifer) (Kungvankij et al., 1986) and other carnivores (Chittino, 1972), and meat of pearl oyster and silkworm pupae for black porgy (Mylio macrocephalus) (Chen, 1990). Hard-boiled egg yolk formed the main food of larvae and fry of milkfish and mullets (Sarig, 1981, Villaluz et al., 1982).

The different feedstuffs used in the culture of brackishwater finfishes varied from place to place depending on the availability of feed ingredients. As cereal grains were cheap in most of the South East Asian countries, the cereal grains and their by-products were used for feeding fishes . Commonly used feedstuffs were roasted rice or wheat flour, broken rice and wheat, maize bran and germ meal, millet, mill sweepings, bran, sago waste and other cereal grains (Huet, 1975, Bardach et al., 1972, Korringa, 1976, Devendra, 1988, Chiu, 1988). Tan et al. (1984) report that there is no clear-cut feed technology for milkfish and a few who practice supplementary feeding use single ingredients like rice bran and bread crumbs. Villaluz et al. (1982) report rice bran as a valuable feedstuff commonly used as supplementary feed for milkfish. Thomforde (1987) observed that supplementary feed of milkfish and tilapia containing 56% rice hulls gave a food conversion of 1.67. Wheat, other cereal grains and sorghum are the important feedstuffs used in Israel for the rearing of mullet (Yashouv and Ben-Shachar, 1967). In Hongkong Mugil sp. fed with rice bran and peanut cake gave a production of 3500 Kg/ha with a food conversion value of 1 1.6 (Sarig, 1981).

Oilcakes have also contributed to the feed ingredients in the culture of many fishes. Soyabean meal or cake and peanut meal are used for rearing of mullets in Taiwan and Hongkong, while groundnut oilcake is the main ingredient used for supplementary feeding in India (Lin, 1967) Groundnut oilcake (66 6%) mixed with rice bran (33 3%) is the usual supplementary feed given to brackishwater finfishes in India (Seghal and Sharma, 1991) Other oilcakes used in Asian countries include castor seed cake, coconut cake, cotton seed cake, palm kernel, sal seed cake, and gingelly oilcake (Devendra, 1988 and Paulraj, 1987).

Artificial diets, compounded from various feedstuffs, that were meant for other fish species and animals have also been used in the culture of brackishwater finfishes. Fish pellets (37 4% crude protein) considerably increased fish production in milkfish ponds compared to lab-lab or plankton as the main food (Fortes, 1984) Commercial chicken starter diets, containing 21% crude protein, used as supplemental diet for milkfish did not significantly increase growth, survival and production of milkfish in fertilized ponds (Otobushin and Lim, 1985) In Queensland, sea bass (<u>Lates calcarifer</u>) was weaned on a commercial salmon starter diet of fine granules containing 52% protein, and 16% fat (Mackinnon, 1986) Among other sources of dry and fresh aquatic weed (<u>Ceratophyllum demersum</u>), concentrated pig blood, fish meal, cassava starch and chicken pellot mixture, only fish fed on chicken pellet gave best growth rate in <u>Oreochromis niloticus</u> (Chiayvareesajja <u>et al</u>, 1987).

2.4 Protein Sources used in Supplementary Feeding

The growth potential of fish is influenced to a great extent by the food quality (Pandian, 1967). Unlike most domesticated farm animals, majority of fish species currently farmed, include carnivores and herbivores and consequently they require high protein in their diet because of low energy requirements (Pandian and Vivekanandan, 1985) This is best exemplified in the experiments of milkfish by Sumagaysay et al.(1991) Feeding milkfish with pelleted diets containing 22% and 27.4% protein resulted in 35.3% and 46.7% higher net profits respectively, whereas feeding with rice bran (containing 11.3% protein) resulted in 34.4% lower net profit from brackishwater ponds. So the main protein source selected for fish feed must have high protein content especially at the younger stages Although, protein is the costliest item in fish feeds (Wee, 1988), various animal and plant sources have been used alone or in combination in fish feeds A brief review of various protein sources used in fish feed formulation is dealt with.

2.4.1 Protein sources of Animal origin.

It is well known, that fishmeal forms the principal source of protein in commercial fish diets. It is also a rich source of energy and minerals, and is highly digestible and palatable for most fishes (Lovell, 1989).

Timbol, as early as 1969, found that in young C chanos, a high protein trout feed based on fishmeal gives a higher weight gain and survival rate than a diet with low protein rabbit-feed containing basically alfalfa Milkfish fry fed on four formulated dry diets containing 40% crude protein with fish meal as the major source of protein, had significantly higher survival and weight gain, compared to those fed on Moina and blended water hyacinth leaves (Santiago et al., 1983). A feed containing 60% fish meal and 16 9% fat was found to be the best diet among a series of dry diets tested for Lates calcarifer fry. However, a diet containing 20% fish meal and 13.4% fat gave practically the same result (Tucker et al., 1988). But in milkfish fry a fish meal based diet containing 30% protein produced better growth than with lower protein levels (Seneriches and Chiu, 1988). Tubongbauna - Marasigne (1990) found that feed from Japanese fish meal based diets was superior to a combination of pasteurized trash fish and Japanese fish meal (1 1 ratio) or pasteurized fish meal alone or local fish meal based diets in Lates calcarifer fry.

However, in the past two decades, the replacement of fish meal protein with other alternative sources was clearly back in focus with increased pressure on diet cost (Smith, 1990) and inconsistent supply. Alternate protein sources which are cheap and locally available and can supplement fish meal without significantly reducing the production, have been put to use Blood-meal-ruminant content mixture is used as a partial or complete substitute for fish meal in commercial catfish rations (Reece et al , 1975) Asgaard (1984) found that slaughter blood had no negative effect on growth, health or organoleptic characters of salmonids. Viola (1975) stated that the meal made from poultry meat and feathers can serve as a good substitute for fish meal in carp diets. However, Kerns and Roelofs (1977) found that the growth rate and feed conversion efficiency were inversely related to the level of poultry waste in common carp diets. Kumar and Singh (1983) noted that pelleted poultry litter resulted in faster growth rate in common carp under laboratory conditions than with traditional feed mi ture of groundnut oilcake and rice bran. The importance of processed piggery waste as feed material for common carp has been reported by Watson (1985). Bull et al (1988) found that slaughter-house waste, vegetable waste, poultry farm waste and press cake waste to be equally effective in the feed of common carp. The percentage weight gain, PER, SGR, and feed conversion efficiencies were not significantly different among the diets

The value of shrimp by-product meal as an alternate protein source in the diets of channel catfish and Liza parsia has been elucidated by Robinette and Dearing (1978) and Kiron (1989) respectively. However, the results indicated poor performance, the reason attributed was that shrimp by-product meal was neither digestible nor palatable as fish meal or that it was deficient in some unidentified factors. Contrary to this, Afolabi et al. (1980) reported better protein efficiency ratio when shrimp and fish wastes were used. Lukowicz (1978) investigated the possibility of replacing fishmeal in carp diets with krill (Euphausia superba) meal, it compared favourably with that of fish meal. When squid was used as food for salmonids and rainbow trout increase in growth was observed (Asgaard, 1984) Squid meal has also been used successfully to replace rice bran in shrimp feeds in Asia (Devendra, 1988). Squid, shrimp and mussel meat extracts are known to be good sources of feeding stimulants, especially for carnivorous fishes like eel, red sea bream and sea bass (Paulraj, 1989).

Jeyachandran and Paulraj (1976, 1977) have shown that silkworm pupae and prawn waste can be profitably utilized as feed for common carp. Defatted silkworm pupae was a better source than non-defatted one in the diet of common carp, with a feed conversion of 2.96 (Nandeesha et al., 1989, 1990).

Use of terrestrial snails in the feed of Oreochromis mossambicus fingerlings has been reported by Shafiei and Costa (1989). In this fish, flesh of snail Achatina fulica produced higher growth rate than chicken feed.

2.4.2 Protein sources of Plant origin.

Various locally available sources of plant proteins have also been incorporated in fish diets instead of fish meal. Among these soyabean meal appears to have been used widely as it is the dominant oil seed protein available world wide and economically viable too (Anon, 1978). It has one of the best amino acid profiles of all protein rich plant feedstuffs, meeting the EAA requirements of fish (N.R.C., 1983). Preliminary results of its use in carp diets have been conflicting, as several workers have reported a reduction in both growth and feed conversion efficiency when higher levels of soyabean meal are included at the expense of fish meal (Viola, 1975, Hepher *et al.*, 1979). However, its successful use as complete replacement of fish meal has been reported in *Oreochromis aureus* (Davis and Stickney, 1978), and a replacement of 25 - 75% dietary protein in tilapia (Jackson *et al.*, 1982, Viola and Arieli, 1983, Davies *et al.*, 1989), and *C.chanos* (Shiau *et al.*, 1988).

Solvent extracted cotton seed can replace 20 - 35%, rape seed 28 - 42%, and sunflower 70% of fish meal in the diets of *Orechromis mossambicus*, *O.niloticus* and *C.carpio* (Dabrowski and Kozlowska, 1981, Viola *et al.*, 1981, 1983, Jackson *et al.*, 1982, Davies *et al.*, 1989, Shiau *et al*, 1990). Full-fat soyabean meal can replace 58% of the diet and cotton seed meal 21% of the fish meal in the diet of *Tilapia nilotica* without significant decrease in growth rate (Lovell, 1980, Viola *et al.*, 1982, Wee and Shu, 1989, Shiau *et al.*, 1990). Roasted Indian mustard seed cake replaced upto 20% fish meal and groundnut expeller cake upto 17% of fish meal in the diet of *Cyprinus carpio* (Jackson *et al.*, 1982) Autoclaved mustard oil cake improved growth performance and food utilization in carp (Hossain and Jauncey, 1990)

A base line information on the potential use of legumes viz, pigeon pea (*Cajanus cajan*), mungo (*Phaseolus radiatus*), kidney bean (*P vulgaris*) and soyabean (*Glycine max*) as protein sources for *C.chanos* is given by De la Pena *et al.* (1987) Of these, only mungo and soyabean at 25% of dietary protein level could replace fish meal to reduce cost without affecting growth, survival and efficiency of feed conversion. Martinez-Palacois *et al* (1988) and Desilva and Gunasekhera (1989) have tried jack bean (Canvalia ensiformis) and green pea (Phaseolus aureus) respectively, in tilapia feeds.

Dried powder of Nymphoides and Spirodella mixed with rice bran has been found to be useful in rearing the fry of carps (Patnaik and Das, 1979). Water hyacinth (Eichhornia crassipes) has been successfully used in the feeds of O niloticus (Edwards et al., 1985). Variable results were obtained when Azolla pinnata was fed to Nile tilapia (SEAFDEC, 1984, Alamazan et al., 1986). A plant mixture of Ceratophyllum demersum, Eichhornia crassipes and Eleocharis ochrostachys pellets has a potential as partial replacement of fish meal (Teshima et al., 1990).

The reports on the use of ipil-ipil (Leucaena leucocephala) leaf meal for rearing tilapia to marketable size are found varying (Camacho and Dureza, 1977, Pantastico and Baldia, 1980, Wee and Wang, 1987, Olvera-Novao et al., 1990) Cassava (Manihot esculenta) leaf meal has been demonstrated to be a viable partial dietary protein source for Nile tilapia (Cruz and Fabian, 1980, Ng and Wee, 1989).

Jackson et al. (1982) have shown that the use of a combination of several plant protein sources is more advisable than as single form, as the protein sources have different limiting amino acids Their experiments with Nile tilapia indicate that cotton seed, rape seed and sunflower seed promoted reasonable growth when provided at 50% of total dietary protein, while copra, soyabean and groundnut might have performed more favourably in the diets, had they been supplemented with single limiting amino acids.

2.4.3 Mixed Protein sources.

For fast growing animals, the protein which contain EAA, in the same balance as those found in the body protein of growing animal is evaluated as high in protein quality (Nose, 1979). However, neither the animal source nor the plant source of protein can provide all the EAA in adequate levels. So usually a mixture of protein sources are used in the formulation of fish feeds

Mitra and Das (1965) evaluated various protein sources for Indian major and minor carp fry. Among the several sources tried, higher survival and yield of carp spawn were obtained when fed with til oil cake, rice powder and black gram, silkworm pupae and fish meal as compared to ricebran.

Several artificial diets compounded from natural and synthetic materials have been tried for the larvae and fry of mullets with limited success (Nash and Shehadeh, 1980). Rangaswamy (1984) obtained best growth of Liza parsia fry with a mixture of bengal gram, prawnhead waste and sago in 2 2 1 ratio. A mixture of wheat middling, cotton seed meal, soyabean meal and tuna fish meal in 4 1 1 1 ratio along with propylene glycol and vitamins is recommended for the rearing of larvae and fry of mullets by Nash and Shehadeh (1980). Studies have shown that feeding Liza parsia fry with wheat flour and fish meal in 1 1 ratio (Chakraborthy et al , 1981) or ricebran and prawn meal in

1 2 ratio (Chakraborthy et al., 1984) gave better results in fertilized ponds than conventional feeds consisting of groundnut oil cake, fish meal or ricebran mixture. Kiron (1989) found that a feed compounded from groundnut oil cake, gingelly oil cake, coconut cake, rice bran, mangrove leaves, fish meal and prawnhead waste providing a dietary protein level of 35% to be the best food for Liza parsia in brackishwater ponds.

Samsi (1979) attempted various feedstuffs like fish meal, meat and bone meal, shrimphead meal, copra meal and ipil-ipil meal as protein sources for milkfish fingerlings. Animal protein sources were better utilized than plant sources Carreon et al (1984) report that growth of milkfish fry supplied with natural plankton were low than those reared on artificial detritus made from rice straw, hulls and chicken manure. Diets with fish meal as major protein source (21%) and the balance protein (16%) level supplemented with animal (shrimp head meal and/or meat and bone meal) and plant (soyabean meal and/or corn gluten meal) sources in a 41% protein diet promoted good growth and survival of milkfish fry Full (8%) or partial (4%) replacement of protein from shrimphead meal with meat and bone meal did not affect the growth of fry significantly However, diet containing shrimphead meal and soyabean meal or shrimphead meal and corn gluten meal or soyabean meal and meat and bone meal is recommended for milkfish fry from a practical standpoint (Alava and Lim, 1988). A formulated diet with fish meal 56 6%, soyabean meal 4 4%, shrimp meal 9%, rice bran 12 7%, cod liver oil 2 7%, corn oil 2.5%, starch 1%, vitamin mixture 0.7% and mineral mixture 3 6% was found to be superior to a diet combination of Oscillatoria and the formulated feed or Spirulina and the formulated diets (Santiago et al., 1989).

In Siganus canaliculatus, a brackishwater herbivore, feed formulated from fish meal , soyabean meal, rice bran, corn meal, coconut oilcake, sea weed gum, fish oil and vitamin premixes was a better source than broiler chicken feed or broiler chicken feed-fish meal combination (Kungvankij et al., 1990)

2.4.4 Unconventional Protein sources.

The use of unconventional protein sources in fish feeds has been reviewed by Jauncey (1982), Tacon and Jackson (1985) ; Wee (1988) and Pantastico (1988).

Atack et al. (1979) used a variety of novel proteins (herring meal, methanophilic bacterium, casein, petroleum yeast and soyabean protein) as the sole source of protein in carp diets. Further studies along this line have indicated that Single Cell Proteins (SCP), especially alkane/petrochemical SCP Candia lipolytica can replace 25-50% (equivalent to a diet yeast SCP inclusion level of 15-30% by weight) and the bacterial SCP Methylophilus methylotrophus, 75% of fish meal in salmonid rations (Tacon and Jackson, 1985 Wee, 1988)

In general, dried algal SCP has only a lower feed value for fish than yeast SCP, bacterial SCP or fish meal (Matty and Smith, 1978, Atack and Matty, 1979). However the studies of Appler and

(Wee *et al.*, 1989). However a negative effect of silage on growth has been reported in *Scophthalmus maximus* by Calcedo-Juanes (1989).

Recently live maggots are used as protein replacements (20% of total dietary protein) in the feeds of *Oreochromis mossambicus* fingerlings (Abalos *et al.*, 1990). Kandasami and Paulraj (1990) have also reported the use of hide fleshings meal (protein meal isolated from hide fleshings) fortified with methionine as an ideal unconventional protein source for *Liza macrolepis*.

The use of leaf protein concentrate (LPC), potato protein concentrate (PPC), and protein hydrolysates as unconventional protein sources in fish feeds are yet to be tested in warmwater finfishes (Tacon and Jackson, 1985).

2.5 Studies on O:N Ratio

The effect of feeding on the magnitude of post-prandial increase in oxygen consumption has been investigated in fishes by Miur and Niimi (1972), Tandler and Beamish (1979), Kaushik and Dabrowski (1983b) and Volssa (1986). The higher the proportion of dietary protein, higher the post-prandial oxygen consumption (Jobling and Davies, 1980). So post-prandial increase in oxygen could be used for comparing nutritive value of different diet formulations (Jobling, 1981). The maximum rate of oxygen consumption of *Lithognathus mormyrus* and *L. lithognathus* were 1.22 and 5.65 times, the routine value of teleost starved for 40 and 152 hrs respectively (Volssa, 1986).

Similarly, one of the end products of nitrogen metabolism, ammonia, could also be used to measure the efficiency of dietary protein utilization (Eggum, 1970, Garcia et al , 1981) A higher rate of ammonia excretion indicate protein of poor amino acid balance (Lovell, 1989). The variation in ammonia excretion may also be influenced by the thermal history of the species (Savitz, 1969).

Reports on the O N ratio are scarce in fishes. However, reports on the post-prandial energy and nitrogen metabolism in the early life history of fish and on the fate of dietary ingredients are available in marine fish larvae (Buckley and Dillman, 1982, Houde and Schekter, 1983, Volssa, 1986) and in some fresh water fishes (Kaushik and Dabrowski, 1983 a, b, Kaushik et al., 1985). Dabrowski and Kaushik (1984) report that in warmwater fish larvae, the post-prandial ammonia excretion increases to more than two fold while oxygen consumption rate triples According to Dall and Smith (1986) complete oxidation of a substrate comprised only of protein, gives a theoretical value of 7 1, while mixed substrates give higher O N ratios. A decreased O N ratio indicates an increased protein catabolism (Capuzzo and Lancaster, 1979) which indicate poor utilization of protein for growth of fish.

2.6 Studies on Optimum Food Ration

The determination of optimum food ration is of great significance in aquaculture in terms of reducing the cost of feeding, maintenance of water quality and growth performance of fish. A common method of calculating daily food ration is on the basis of percentage of live body weight of fish (Hepher, 1988)

The influence of rate of feeding on growth rate, conversion efficiency, body composition and metabolism have been extensively studied by Pandian (1967) on Megalops cyprinoides and Ophiocephalus striatus, Pandian and Reghuvaran (1972) on Tilapia mossambica, Chua and Teng (1982) on Epinephelus salmoides, Macintosh and Desilva (1984) on O.mossambicus and hybrid of O niloticus x O aureus, Teshima et al (1984) and Chiu et al (1987) on C chanos and Karmakar and Ghosh (1984) and Kiron and Paulraj (1988) on Liza parsia

The variations in the daily consumption of food have been reported in fishes due to size and/or age (Desilva and Perera, 1983 Desilva et al, 1986), salinity (Desilva and Perera, 1983), and temperature (Boehlert and Yoklavich, 1983) variations.

Pandian and Reghuvaran (1972) reported that an average quantity of 35 mg/ g of fish/day is the optimum food ration for O mossambicus with Tubifex tubifex as feed In estuarine grouper (Epinephelus salmoides), optimum food ration was at 5% of the body weight, though feeding at 9% body weight gave the maximum growth rate (Chau and Teng, 1982) The best food conversion of O mossambicus fry was at 24% of body weight, while it was at 12% of body weight for the hybrid fry of O niloticus and O aureus (Macintosh and Desilva, 1984) Milkfish (C chanos) fingerlings showed best growth when reared on a purified diet containing 35% casein and 15% gelatin as protein sources at 30-50% of body weight, twice daily (Teshima et al, 1984) The specific growth rate of *O.mossambicus* fry (of size 0.8760-1.0810 g) was the highest at 4% feeding level. However, the same fish of still larger size (3 0374-3.3609 g), required a feeding rate of only 3% of the body weight for producing the highest SGR (Desliva *et al.*, 1986) According to Chiu *et al.* (1987) an increase in the feeding level from 5 to 9% of body weight gave a significant increase in the weight gain of *C chanos* juveniles. In *Liza parsia* fry, the food conversion efficiency was best at 4% feeding level (Kiron and Paulraj, 1988)

Various reports are available on the optimum feeding level practised in brackishwater pond culture . The feeding level of yellow tail cultured in Japan is reported to be at a rate of 10% of body weight in grow-out, the feed being moist fish meal and corn gluten (Milne, 1972). In Italy, mullet juveniles are fed at a rate of 5-7% of their body weight, with commercial diets (Korringa, 1976). In Thailand, the nursery rearing of *Lates calcarifer* is done at a feeding level of 100% of their body weight during the first week of rearing by the second week ration is reduced to 60% of body weight and then to 40% of body weight by third week, using chopped ground trash fish. Similarly in the grow-out ponds, they are fed with chopped trash fish at a rate of 10% of body weight for the first two months and then reduced to 5% of body weight (Kungvankij *et al.*, 1986).

MATERIALS AND METHODS

3 MATERIALS AND METHODS

Experiments were conducted to evaluate the efficiency and keeping quality of five feeds compounded from five different protein sources, for the nursery rearing of C chanos fry The optimum food ration of the selected feed, which gave best growth performance in the first experiment was also studied

3.1 Preparation of Feeds

3.1.1 Feed Ingredients.

The different protein sources used in the study were as follows.

Animal origin	Fish meal squid meal, clam meal prawn head waste meal
Plant origin	Soyabean meal.

Other ingredients in the compounded feeds were groundnut oil cake, tapioca powder, cod liver oil vitamin and mineral mixtures

Fish meal and squid meal were prepared by boiling and pressing fish (Nemipterus sp.) and squid respectively, to remove their oil content These were then sundried and ground well Clam meal and prawnhead waste meal were prepared by sundrying and powdering, clam meat and prawnhead waste respectively Fish were bought from the Cochin

Fisheries Harbour Thoppumpady, squid from 'Toyo Fisheries, Kochangadi', clam meat from local market and prawnhead waste from a local peeling shed. Soyabean meal used in the study was of 'HIMEDIA' make. Cod liver oil was obtained from commercial market. Mineral mixture was of USP XIV (SISCO RESEARCH LABORATORIES PVT. LTD., BOMBAY). Vitamin mixture (SUPPLEVITE - M) was obtained from the Biochemistry Department of College of Fisheries. 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.

For estimating the moisture level Boyd's (1979) method was used. The sample was heated to 105°C for 30 minutes and then dried at 65°C till a constant weight was obtained. The crude protein content was estimated by Microkjeldahl's method (AOAC, 1975). The nitrogen content was then 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. Method of Pearson (1976) was used to estimate the crude fibre. The ash content was estimated by burning the sample at 550°C±10°C for 6 hrs in a muffle furnace. The carbohydrate content was found out by Hasting's (1976) difference method as Nitrogen Free Extract (NFE).

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 preparation of test diets.

Test diets were formulated by keeping their protein level constant at 40%. They were

1. FM (Feed with fish meal as the chief protein source)
2. SQM (Feed with squid meal as the chief protein source).
3. CM (Feed with clam meal as the chief protein source)
4. PHM (Feed with prawnhead waste meal as the chief protein source)
5. SYM (Feed with soyabean meal as the chief protein source).

The proportion of various ingredients used for the formulation of pelleted feeds and their contribution to the total dietary protein are given in Table 1. The ingredients (except vitamin and mineral mixture, and cod liver oil) for each feed were mixed thoroughly, Each feed was then hand kneaded using water (1:1.25 w/v) to get a soft dough. It was autoclaved at ambient pressure for 30 minutes, cooled, and vitamin mixture dissolved in cod liver oil and mineral mixture were added. Then the dough was again mixed thoroughly. The dough was then pelletized and dried in a hot air oven at 60°C, till the moisture content was less than 10% (overnight). The pellets were then ground and passed through No.40 sieve (425 μm). They were then packed separately in plastic bags and stored free from moisture and sunlight.

3.1.4 Proximate composition of the Formulated Feeds.

Immediately after the preparation of the feeds, their proximate composition were analysed, using the same methodology adopted for the ingredients. The proximate analysis was also carried out at the end of 4 months of storage period.

The energy value of each feed was calculated in Kcal/g by multiplying factors 8.5 for fat, 3.5 for carbohydrate and 4 5 for proteins, as adopted for *Oreochromis mossambicus* by Jauncey (1982).

3.2 Sinking rate of Feed Crumbles

A pinch of each of the dried ground feed was placed in an aquarium measuring 1 25 x 0.5 x 0 5 m and the time taken by them to traverse the water column was recorded using a stop watch This was tried 4 times for each feed, and the average sinking rate was recorded in cm/sec. Sinking rate was measured soon after the feed preparation and also after 4 months of storage

3.3 Water Stability of Formulated Feeds

Water stability of the feeds were determined using the method of Jayaram and Shetty (1981). It was found out by determining the percentage dry matter recovered after exposing the pellets in water for 3 hrs

3.4 Microbiological quality of Formulated Feeds

The microbiological quality of the feeds were assessed by determining the total heterotrophic bacterial and fungal population Total viable bacterial count was determined using the Plate Count Agar (Tryptone 0 5%, Yeast extract 0 25%, Dextrose 0 1%, Agar 1 5%, pH 7±0 2) by pour plate method (Seeley and Van Wemark, 1975) For estimating the total viable fungal count Sabaraud Dextrose Agar (Mycological peptone 1%, Dextrose 4%, Agar 1.5%, pH 5 6 ± 0 2) was used in the same way (Goss, 1972)

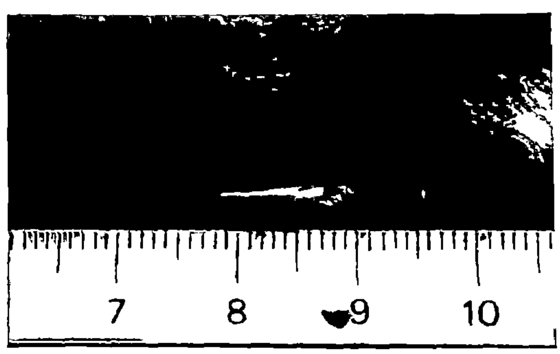
3.5 Keeping quality of Formulated Feeds

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

3.6 Experimental Milkfish fry and their Acclimatization

Milkfish fry of nearly four weeks old and with an average size of 1.4 cm/0014 g were obtained from the Fisheries Station Puduveypu of Kerala Agricultural University They were transported to the College of Fisheries in oxygen filled plastic bags in water of 25 ppt salinity The fry were then introduced into oval fibre glass tank of 1 ton capacity. After temperature acclimatization, they were acclimatized to

Plate I



a final salinity of 5 ppt by gradually replacing the sea water with freshwater The whole process took 3 hrs The fry were then fed with groundnut oil cake, twice daily, till the start of the experiment Aeration was provided throughout acclimation period. Uniform sized, healthy fry (See Plate I) alone were selected for the experimental purpose

3.7 Experimental tanks and Water quality management

Plastic tanks (60 x 40 x 40 cms) of 50 litres capacity were used to carry out the experiment Aeration was provided to the tanks with the help of an air compressor using airstones (See Plate I I).

Experiments were conducted in brackishwater of 5 ppt salinity, which was filtered twice using bolting silk before filling the tanks Everyday, one third of the water was exchanged with water of same salinity Complete water of the tanks were replaced once in every week

3.8 Study to Evaluate the Supplementary Feeds

Healthy milkfish fry of average total length (1 79 cm) and weight (0 017 g) were used for the experiment (See Plate I) They were starved for 24 hrs prior to the experiment The total length was measured to the nearest centimetre from tip of snout to the tip of tail The fry were then blotted dry carefully between folds of filter paper and weighed in an electronic balance to the nearest milligram About 50 fry were weighed and dried in an oven at 40°C for 48 hrs, to carry out the initial biochemical composition of the fish

Plate II

Eight numbers of C chanos fry were then introduced to each tank They were given any one of the five formulated feeds Each feed was replicated four times Thus there were 20 tanks 4 tanks for each of the five feeds

The fry were fed, with the experimental diets ad libitum twice daily, in petridishes kept at the bottom of the tanks Feeding was done in the morning and the evening The left over feed was recovered, before feeding next time The feed-remains were separated from the salt adhering to it and dried in an oven at 60°C to a constant weight

3.8.1 Determination of Digestibility co-efficient.

Digestibility co-efficient was determined using the method described by Halver (1989) Faeces were collected, for determining the digestibility co-efficient studies, from the second week onwards for the next 20 days A large volume bulbous siphon was used for the faeces collection The faeces along with water was sieved through a bolting silk, rinsed with distilled water to remove the salt adhering to it and then transferred to a pre-weighed beaker It was then dried in an oven at 60°C to a constant weight The dried faecal matter was homogenised before analysing for protein and lipid

3.8.2 Monitoring of Physico-Chemical parameters of water.

Water temperature of the experimental tanks was monitored daily at 9 a m. using graduated mercury thermometer with an accuracy of 0 1°C and dissolved oxygen content by Winkler's method (Strickland and Parsons, 1968) Salinity was measured twice a week using a refractometer The pH of the water was measured twice a week with universal indicator solution.

3.8.3 Recording of observations.

Parameters like the food conversion ratio, protein efficiency ratio protein and lipid digestibility co-efficients and specific growth rate were calculated using the method described by Hardy (1989)

3.8.3.1 Food Conversion Ratio (FCR)

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

3.8.3.2 Protein Efficiency Ratio (PER)·

$$PER = \frac{\text{Average live weight gain}}{\text{Average protein consumed in dry weight}}$$

3.8.3.3 Protein and Lipid Digestibility co-efficients.

Digestibility co-efficient of any nutrient is calculated as follows

$$DC = \frac{\text{Nutrient digested}}{\text{Nutrient ingested}} \times 100 \text{ or } \frac{(F \times P) - (E \times Q)}{(I \times P)}$$

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

3.8.3.4 Survival rate.

Survival rate was calculated as follows

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

3.8.3.5 Growth.

Growth was calculated both in terms of length and weight, using the formula

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

3.8.3.6 Specific Growth Rate (SGR).

$$SGR = \frac{\log_e W_2 - \log_e W_1}{t_2 - t_1} \times 100$$

where

W_1 = Weight at time t_1, W_2 = Weight at time t_2

It gives the average percentage increase in body weight per day of fish over 42 days

3.8.4 Biochomical analysis of Carcass

Proximate composition of the carcass of fishes was done after 42 days of growth, to determine the best supplementary feed following the standard procedures mentioned earlier

3.8.5 Estimation of O:N ratio.

After feeding the fish for 42 days, they were subjected to test the O N ratio by respirometry. Four samples from each treatment were taken for the study. Initially the fish were acclimatized in the chamber of the respirometer for an hour The chamber was flushed with the same water in which the fish were kept throughout the equilibration After acclimatization, water samples were taken from the respirometer for estimating the initial dissolved oxygen and ammonia content of the water Then the respirometer was closed and placed in a water bath, maintained at a temperature of 30 ± 1°C, for one hour Water samples were then drawn for the estimation of final dissolved oxygen and ammonia content The difference between the initial and final values gives the oxygen consumed and ammonia excreted by the fish during the period of the experiment. After the completion of experiment, the fish were blotted dry in a filter paper and weighed Oxygen was measured using Winkler's method and ammonia by Spectrophotometric method (Strickland and Parsons, 1968). The results were expressed as mg O_2 /g/hr for oxygen consumption and mg NH_3 -N/g/hr for ammonia excretion O N ratios of individual fishes fed with different protein sources were then estimated following the method given by Bayne et al (1985)

3.9 Study on the Optimum Food Ration

The feed which gave the best growth performance and conversion efficiency of the first experiment was selected for the study of optimum food ration. Eight numbers of milkfish fry (2 0 cms/0 107 g) were introduced to each tank They were then fed at 0,3, 6, 9, 12, 15, 18 or 21% of the body weight, twice daily, for 21 days Each treatment was replicated thrice The survival rate, specific growth rate and food conversion ratio were then analysed. Optimum food ration was calculated using the tangent method.

3.10 Statistical Analysis

Analysis of Variance (Snedecor and Cochran, 1968) was carried out for the collected data Rations and percentage value (X) were transformed into Arc sin values ($\sin^{-1}\sqrt{\frac{x}{100}}$) for analysis

RESULTS

4 RESULTS

4.1 Proximate Composition of Feed Ingredients and Formulated Feeds

4.1.1 Feed Ingredients.

The proximate composition of ingredients used in the formulation of experimental diets is given in Table 2. Moisture content of the ingredients varied from 7.61 to 9.77%, the minimum being for squid meal and the maximum for tapioca powder Squid meal had the highest crude protein content (80.46%), followed by fish meal (70.22%). The crude protein content of other ingredients was clam meal - 50.62%, prawnhead waste 50.15%, soyabean meal 47.6%, groundnut oil cake - 34.88% and tapioca powder 6.92%. The crude fat content of the ingredients varied from 0.9 to 11.1%. Clam meal had the highest fat content and tapioca the lowest. The crude fat content of other ingredients viz., groudnut oil cake, prawnhead waste, squid meal, fish meal and soyabean meal was 7.06, 6.9, 6.5, 5.9 and 2.4% respectively The crude fibre content of the ingredients varied from 0.7% (fish meal) to 8.6% (soyabean meal). Tapioca powder had the highest NFE content (72.1%), while squid meal had the lowest (0.6%). The ash content was maximum for prawnhead waste (23.65%), which was followed by fish meal (14 2%). The minimum content of ash was found in squid meal (4.03%).

4.1.2 Formulated Feeds.

The proximate composition of the formulated feeds is given in

Table 2. Proximate composition of the ingredients used in feed formulation * (% dry weight)

Ingredients	Moisture	Crude protein	Crude fat	Crude fibre	Carbohydrate (N-Free Extract) NFE	Ash
Fish meal	8.08	70.22	5.90	0.70	0.90	14.20
Squid meal	7.61	80.46	6.50	0.80	0.60	4.03
Clam meal	9.65	50.62	11.10	4.11	16.90	7.62
Prawnhead waste	7.88	50.15	6.90	6.53	4.89	23.65
Soyabean meal	8.94	47.60	2.40	8.60	25.36	7.10
Groundnut oil-cake	9.43	34.88	7.06	7.50	33.80	7.33
Tapioca	9.77	6.92	0.90	4.30	72.10	6.01

* Average of four values

Table 3. The moisture content of the feeds varied from 8.5 to 9.6%. The crude protein content of all the feeds was around 40% (39.78 to 40.44). The crude fat content was highest for CM (8.1%) and the lowest for SYM (5.8%), while it was 7.2, 7.1 and 6.5 for PHM, SQM and FM respectively. The fat content seems to have an inverse relationship with moisture content. The crude fibre content was the highest in SYM (8.8%) and the lowest in FM (3.59%). The NFE content of PHM was 20.6% and CM 24.3%. For all the other feeds it was around 26% The ash content of the feeds varied from 9.75 to 17.04-highest being for PHM and the lowest for SYM. The feeds were almost isocalorific and their energy content varied from 3.13-3.33 Kcal/g.

Effect of storage on the keeping quality of the feeds, as indicated by various parameters is given in Table 4. Moisture and the NFE showed a decrease in their contents after four months of storage (Fig. 1). The highest percentage increase in the moisture content was for SYM. Feeds FM, PHM, SQM and CM showed an increase in moisture content of 15.03, 14.82, 14.61 and 11.06%, respectively. The increment in the NFE content in feeds after four months of storage was in the order of PHM (20 98%), CM, (15%), SYM (8.27%) and FM (4.91%). The drop in the crude protein content was 5.2% for PHM while it was 4% for CM. For other diets it was between 2 to 2.7%. The crude fat content of feeds

Table 3. Proximate composition of formulated feeds * (% dry weight)

Parameters %	Diets				
	FM	SQM	CM	PHM	SYM
Moisture	9.45	9.38	8.50	8.60	9 60
Crude protein	40.28	40 44	39.78	39.86	40 10
Crude fat	6.50	7.10	8.10	7.20	5.80
Crude fibre	3.59	4.90	5.82	6.70	8.80
Carbohydrate (NFE)	26.70	26.00	24.30	20.60	26 00
Ash	13.48	12.18	13.50	17.04	9.70
Energy content	3.30	3.33	3.33	3.13	3 21

* Average of four values

Table 4. Effect of storage in the keeping quality of formulated feeds.

Parameters	FM			SQM			CM			PHM			SYM		
	Fresh	After 4 months storage	% age difference	Fresh	After 4 months storage	% age difference	Fresh	After 4 months storage	% age difference	Fresh	After 4 months storage	% age difference	Fresh	After 4 months storage	% age difference
Average sinking rate cm/sec	2 80	3 90	-	2.40	3 40	-	1.20	1 90	-	1.70	2 60	-	3 40	4.50	-
Stability %	86 70	84 09	3.01	86 8	85.52	2 93	88 10	85 63	2 80	78.90	75 10	4 82	87.80	84 78	3 44
Moisture %	9 45	10 87	+15.03	9 38	10 75	+14 61	8 50	9 44	+11 06	8 60	9.88	+14 82	9 60	11.27	+17.4
Crude protein %	40 28	39 41	-2 16	40 44	39 63	-2 00	39 78	38 20	-4 00	39.86	37.80	-5 20	40.10	39.03	-2.7
Crude fat %	6 50	6 25	-3 80	7 1	6 61	-6 90	8 10	7 32	-9 60	7 20	6 68	-7 20	5.80	5 64	-2.8
Ash %	13 48	12 01	-10 91	12 18	9 94	-18 40	13 50	11 80	-12 60	17 04	14.38	-15.61	9 70	7 81	-19.48
Crude fibre %	3 59	3 45	-3 90	4 90	4.71	-3 90	5 82	5 30	-8 90	6 70	6 02	-10 15	8 80	8 10	-7.95
Carbohydrate % (NEF)	26 70	28 01	+4 91	26 00	28.36	+9 10	24 30	27 94	+15 00	20 60	25 24	+20 98	26 00	28 15	+8 27
Fungal count cfu/g	0	3×10^{6}	-	0	5×10^{6}	-	0	$4\ 8 \times 10^{7}$	-	0	8×10^{6}	-	0	$7\ 1 \times 10^{6}$	-
Bacterial count cfu/g	$1\ 48 \times 10^{6}$	$3\ 7 \times 10^{7}$	-	$1\ 1 \times 10^{5}$	$1\ 5\ 10^{7}$	-	$1\ 1 \times 10^{5}$	$3\ 1 \times 10^{7}$	-	$3\ 9 \times 10^{5}$	4.2×10^{7}	-	$3\ 1 \times 10^{5}$	1.8×10^{7}	-

Average of four values

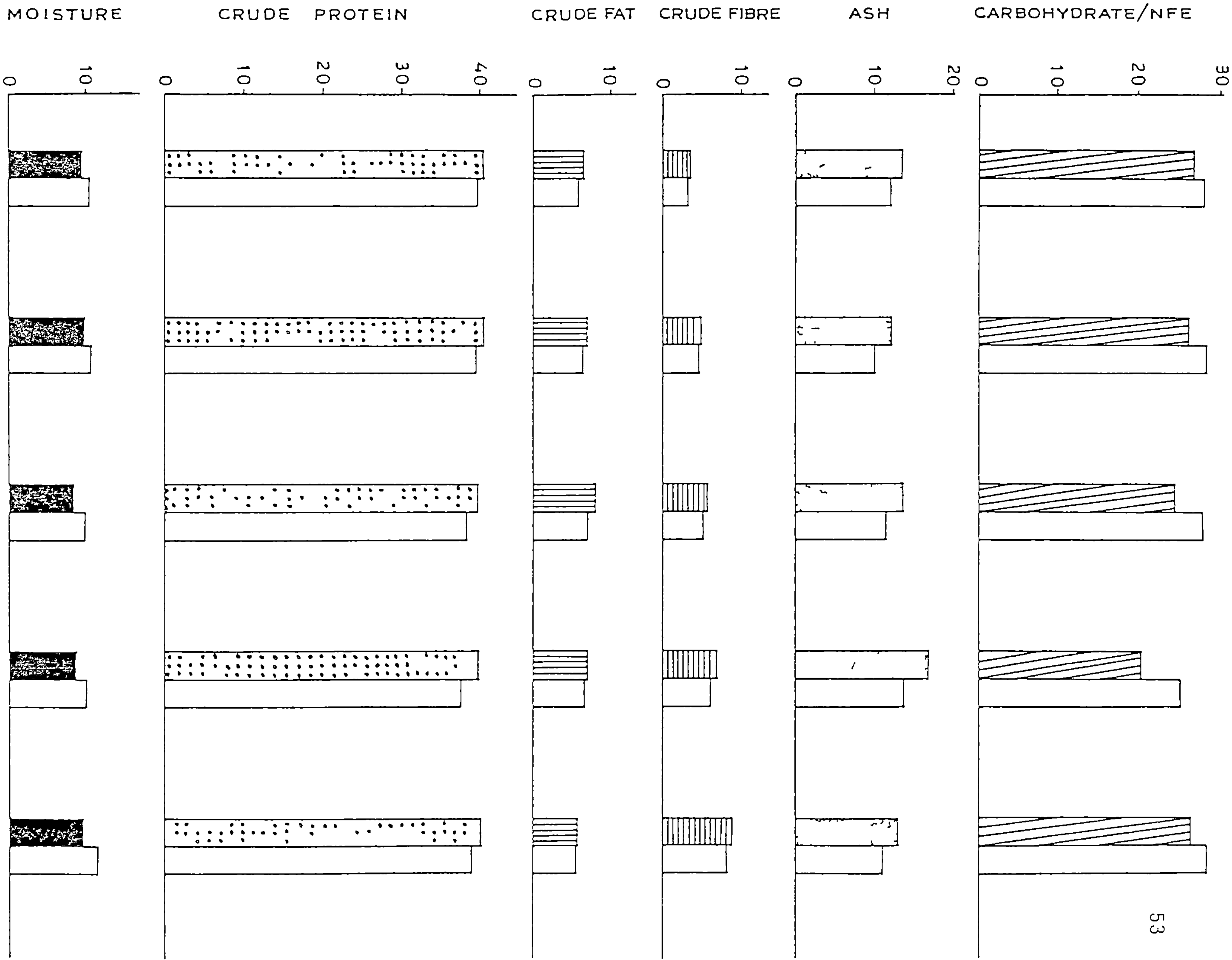
was found to be lowered in the order of CM (9.6%), PHM (7 2%), SQM (6.9%), FM (3.8%) and SYM (2.8%), as in the same order of fat content in the fresh feeds. SYM (19.48%) showed the maximum decrease in the ash content among the feeds, while the minimum decrease was for FM (10.91%). The decrease in the crude fibre content was the highest for PHM (10.15%) and lowest for FM and SQM (3.90%).

4.2 Sinking Rate of Formulated Feed crumbles

The sinking rate of SYM was the highest, both before (3.4 cm/sec) and after storage of 4 months (4.5 cm/sec). The sinking rate of other feeds was 1.7 (PHM), 2.4 (SQM), 2.8 (FM) and 1.2 cm/sec (CM). All the feeds exhibited an increase in the sinking rate after storage (Table 4). The sinking rate after storage was 1.9, 2.6, 3.4, and 2.9 cm/sec for CM, PHM, SQM, and FM respectively.

4.3 Water Stability

The most stable feed at the end of three hours in the water was CM (88.1% dry matter), and the least stable was PHM (78.9%). The initial values of water stability of other feeds were 86.7% (FM), 86.8%(SQM) and 87.8%(SYM). The water-stability of feeds reduced after four months of storage, the range in the values being 75.1%(PHM), and 84.78%(SYM) (Table 4).


MOISTURE
CRUDE PROTEIN
CRUDE FAT
CRUDE FIBRE
ASH
CARBOHYDRATE/NFE
0
10
20
30
40
FM
SQM
CM
PHM
SYM

4.4 Microbiological quality of Formulated Feeds

In the fresh samples there were no fungal colonies But after four months of storage, fungal colonies were found to occur (Table 4). The maximum fungal count was found in CM (4.8×10^7 cfu/g), while it was in the range of $3x10^6$ to $7.1x10^6$ cfu/g for other feeds. The total plate count (bacterial count) in the fresh samples was in the range of $1.1x10^5$ to $3.9x10^5$ cfu/g (Table 4). The highest count was for PHM, followed by FM and SYM. While SQM and CM had the same bacterial count ($1.1x10^5$ cfu/g). There was an increase in the total plate count after four months of storage, the range being $3.6x10^6$ to $3.08x10^6$ cfu/g. The increment in TPC was maximum for CM ($3.08x10^7$).

4.5 Evaluation of Supplementary Feeds

4.5.1 Water quality maintenance.

4.5.1.1 Temperature.

Table 5 gives the range of temperature in the experimental tanks during the study for the evaluation of supplementary feeds. The water temperature ranged from 27.1 to 30.8°C. The mean values of temperature in each treatment during the six weeks of study are also given in Table 5. The temperature did not vary much between the experimental tanks during the study.

Table 5. Range of temperature (in °C) of the experimental tanks during the study for the evaluation of formulated feeds (Values in the paranthesis give the range of temperature)

Treatment	Weeks Mean ± SD					
	1	2	3	4	5	6
FM	27.27±0.17 27.1-27.5	27.9±0.21 27.8-28.1	28.63±0.09 28.6-28.7	29.4±0.08 29.3-29 5	29.98±0.83 29.9-30.1	30.6±0.14 30.5-30.8
SQM	27.22±0.30 27 2-27.5	28.01+0.11 28.0-28.2	28.43±0.05 28.4-28.5	29.4±0 18 29.3-29.5	29.98±0.10 29.6-29.9	30.55+0.17 30 4-30.8
CM	27.13±0.24 27.1-27.4	28.03±0.09 27 9-28 1	28.58±0.05 28 5-28.6	29.43±0.05 29 4-29.5	30 08±0.15 29.9-30.2	30.30±0.10 30.1-30.6
PHM	27.33±0.09 27.2-27.4	28.10±0.14 28.0-28 3	28.53±0.19 28.4-28.8	29 23±0 13 29.1-29 4	30.18±0.05 30.1-30.2	30.38±0.10 30 3-30.5
SYM	27.21±0.27 27.2-27.4	28.1±0.18 27.9-28.3	28.63±0.13 28 5-28.8	29.48±0 10 29.4-29.6	29.48±0.10 30.1-30.3	30.7±0.08 30.6-30.8

4.5.1.2 pH.

The pH of the water fluctuated from 7.6 to 8 3 during the study The mean values of pH and its range in each treatment during the six weeks study are given in the Table 6. There was not much fluctuation in pH between the experimental tanks.

4.5.1.3 Dissolved oxygen.

The dissolved oxygen content of water recorded over the experimental period is shown in Table 7. The value varied from 6 8 to 8.8 ppm. However, there was not much variation in the dissolved oxygen content of water, as aeration was provided throughout the experiment.

4.5.2 Efficiency of Feeds.

4.5.2.1 Food Conversion Ratio.

FCR was best with CM (1.74 ± 0 025) indicating that it was the most efficient feed among the five. The poorest FCR of 10.95 ± 1 54 was given by fish fed on PHM (Table 8) A diet based on SQM could provide a better FCR (2 94) than from FM (3 65) (Fig.2), however their conversion rates were not significantly different

Analysis of Variance shows that FCR of CM was significantly higher ($P \leq 0.01$) than others. FCR of SYM and PHM was also significantly

Table 6. Fluctuations of pH in the experimental tanks during the study for the evaluation of formulated feeds (Values in paranthesis give the range of pH)

Treatment	Weeks means ± SD					
	1	2	3	4	5	6
FM	8.15±0.44 7.6-8.2	8.08±0.28 7.8-8.1	8.13±0.13 8 0-8.2	8.08±0.19 7.8-8.2	8.13±0.13 8.0-8.1	8.15±0.17 8.0-8.1
SQM	8.1±0 14 7.8-8.2	8.13±0.05 8.1-8 2	8.03±0.22 8.0-8.2	8.05±0.13 8.0-8.2	8.05±0.1 8.0-8.1	8 18±0.1 8.1-8.2
CM	8.0±0.10 8.0-8.2	8.05±0.06 8.0-8 1	8.03±0.05 8.0-8.1	8.18±00.05 8.1-8.2	8.0±0.11 8.0-8.2	8.03±0.13 8.0-8.2
PHM	8.23±0.13 8.2-8.3	8.25±0.17 8.2-8 3	8.25±0.17 8.2-8.3	7 93±0 42 7 6-8.2	8.2±0 08 8.2-8.3	8.05±0.05 8.0-8.3
SYM	8.06±0.04 8 0-8.1	8.0±0 07 8.0-8.2	8.12±0.05 8.1-8.2	8 05±0.29 7.6-8 2	8.28±0.1 8.2-8.3	8.08±0.08 8.0-8.2

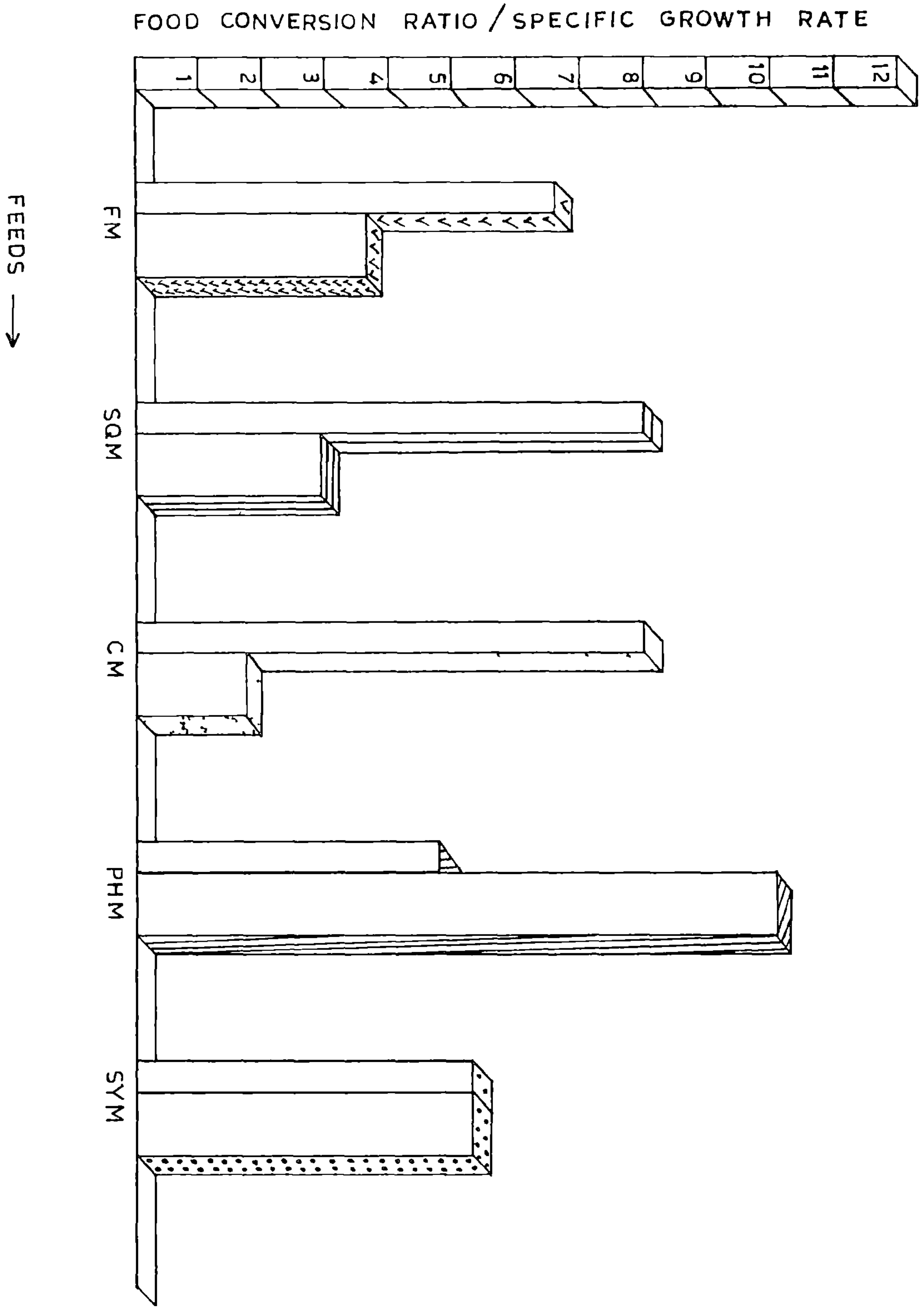
Table 7. Variations in the dissolved oxygen content in the experimental tanks during the study for the evaluation of formulated feeds.

Treatment	Replication	Weeks					
		1	2	3	4	5	6
FM	1	8.4	8.4	6.8	8.0	8.8	8.0
	2	8.0	8.8	7.8	8.2	8.0	8.2
	3	7.2	8.4	7.4	7.8	8.0	8.0
	4	7.8	8.53	7.3	8.0	8.2	8.1
AV+SD		8.16±0.44	8.53±0.16	7.33±0.36	8.0±0.14	8.4±0.32	8.08±0.08
SOM	1	8.0	8.4	8.2	8.8	8.0	8.0
	2	7.6	8.0	7.4	8.6	7.4	7.8
	3	8.2	7.2	6.8	8.8	7.6	7.8
	4	7.9	7.9	7.4	8.7	7.7	7.9
AV+SD		7.93±0.22	7.88±0.43	7.45±0.50	8.3±0.08	7.7±0.25	7.89±0.08
CM	1	8.4	7.2	8.4	7.8	8.2	7.8
	2	7.4	7.6	7.8	7.6	8.0	8.0
	3	7.8	7.7	8.4	8.2	7.6	7.2
	4	7.4	7.4	8.5	8.1	7.9	7.7
AV+SD		7.75±0.41	7.48±0.19	8.23±0.28	7.93±0.24	7.93±0.22	7.68±0.29
PHM	1	8.2	8.3	8.6	8.2	8.0	7.8
	2	7.8	7.8	8.2	7.8	8.6	8.0
	3	7.0	8.0	8.4	8.6	7.4	7.2
	4	7.7	8.2	8.4	8.2	8.0	7.7
AV+SD		7.68±0.43	8.08±0.19	8.4±0.14	8.3±0.24	8.0±0.42	7.68±0.29
SYM	1	7.8	8.2	7.8	8.4	8.0	8.4
	2	8.4	8.1	8.2	8.4	8.2	8.0
	3	7.8	7.9	8.0	8.6	7.8	8.2
	4	8.6	8.3	8.6	8.1	8.0	8.2
AV+SD		8.15±0.36	7.9±0.52	8.15±0.30	8.38±0.18	8.0±0.14	8.2±0.14

* Average of 3 values

Table 8. Food conversion ratio of C.chanos fry fed with various formulated feeds for 42 days

Feed	Replication	Average initial weight (g)	Average final weight (g)	Average live weight gain (g) a	Av. wt. of food consumed (g) b	Food conversion ratio (FCR) b/a	Mean ± SD
	1	0.019	0.312	0.293	1 0243	3.50	
	2	0.011	0.303	0.292	1.0693	3.66	
FM	3	0.022	0.307	0.285	1 0768	3.78	3.65±0.099
	4	0.028	0.307	0.279	1.0208	3.66	
	1	0.011	0 377	0.366	1 0761	2.94	
	2	0 015	0.394	0.379	1.0988	2.90	
SQM	3	0.011	0.375	0 364	1 0801	2.97	2.94±0.025
	4	0.019	0 398	0.380	1 1215	2.95	
	1	0.011	0.456	0.445	0.7831	1.76	
	2	0.018	0 475	0.457	0.7769	1.70	
CM	3	0.019	0.463	0.444	0 7815	1 76	1.74±0 025
	4	0.019	0.471	0.452	0.7915	1.75	
	1	0.014	0.118	0.104	1.1919	11.46	
	2	0.022	0.124	0.102	0.9090	8.91	
PHM	3	0.021	0 133	0.112	1.1545	10.31	10.95±1.54
	4	0.011	0.110	0.099	1 2991	13.12	
	1	0.022	0.155	0.133	0 6954	5.23	
	2	0.011	0.152	0.141	0.8274	5.87	
SYM	3	0.016	0.154	0.138	0.6943	5.03	5.32±0.32
	4	0.019	0.156	0.137	0 7070	5.16	


FOOD CONVERSION RATIO / SPECIFIC GROWTH RATE
1
2
3
4
5
6
7
8
9
10
11
12
FEEDS
FM
SQM
CM
PHM
SYM

different from each other (Table 14)

4.5.2.2 Protein Efficiency Ratio.

PER too, was best with CM (1 45 + 0 021) PER values of various feeds are given in Table 9 SQM and FM had a PER of 0 84 and 0 68 respectively. PER was relatively poor (Fig 3) for SYM (0 47) but the lowest was for PHM (0 23) fed fish

Analysis of Variance shows PER of all feeds to be significantly different from each other (Table 14)

4.5.2.3 Protein Digestibility co-efficient.

Table 10 gives the protein digestibility co-efficient of different feeds in C chanos fry It was the highest for fish fed on CM (97 24 + 0 558) and the lowest for PHM (65 23 ± 0 379) The protein digestibility co-efficient of other feeds were, FM - 86 56 ± 0 024, SQM-91 86 ± 0 172, and SYM 77 78 + 0 097 respectively (Fig 4)

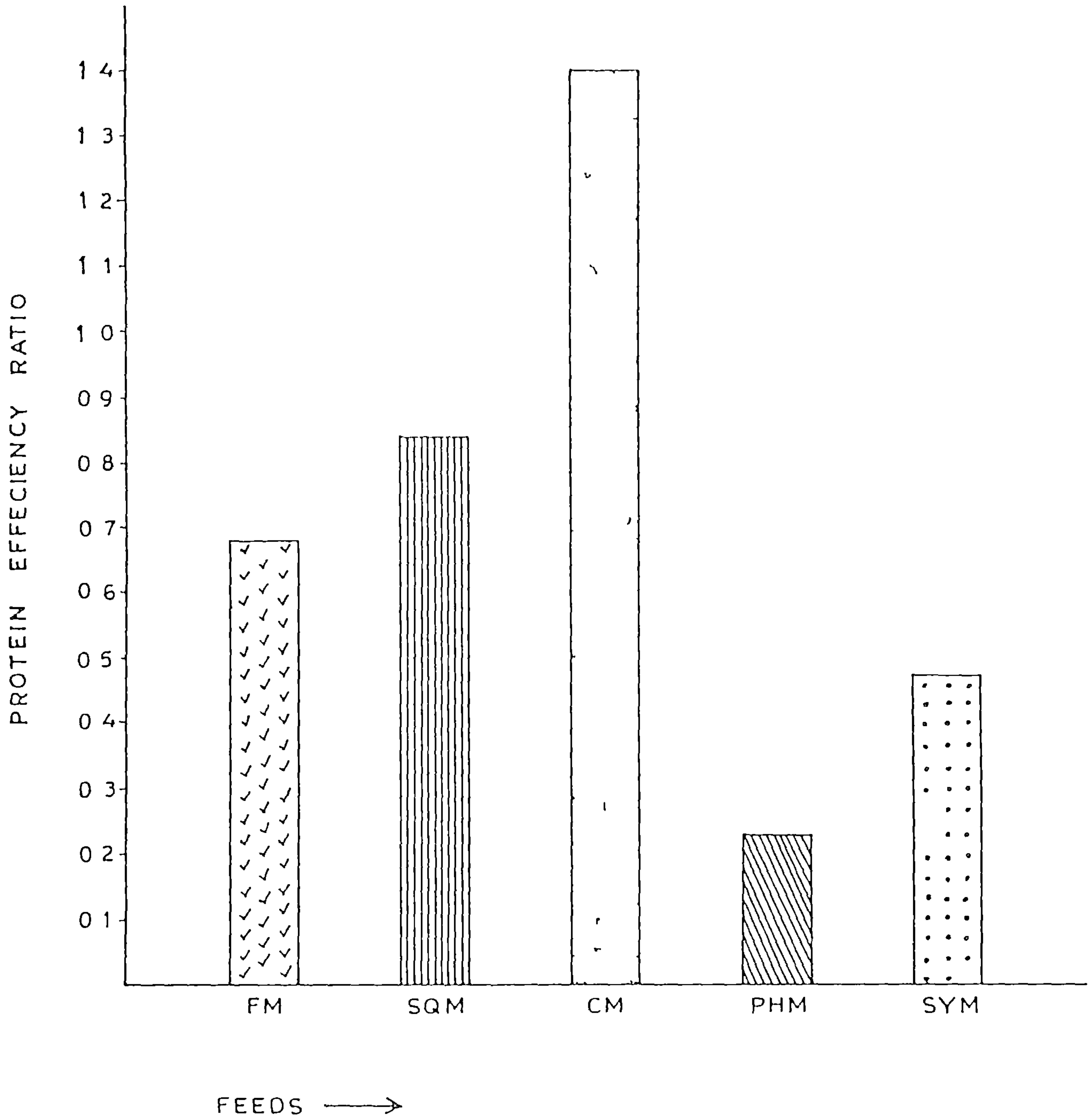
Statistical analysis showed that protein digestibility of all feeds was significantly different from each other (Table 14)

4.5.2.4 Lipid Digestibility co-efficient.

Digestibility of lipid was maximum for fish fed on CM(98 08 ± 0 43%) However digestibility of lipid from SQM was not

Table 9. Protein efficiency ratio of C.chanos fry fed with various supplementary feeds

Replication	Average initial weight (g)	Average final weight (g)	Average live weight gain (g) a	Average weight of food consumed (g)	Average weight of protein consumed (g) b	PER a/b	Mean ± SD
1	0.019	0.312	0.293	1.0243	0.4126	0.71	
2	0.011	0.303	0.292	1.0693	0.4307	0.68	
3	0.022	0 307	0.285	1.0768	0 4347	0.66	0.68±0 018
4	0 028	0 307	0 279	1.0208	0 4112	0.68	
1	0.011	0 377	0 366	1.0761	0 4352	0.84	
2	0.015	0 394	0.379	1.0988	0 4444	0 85	
3	0 011	0.375	0 364	1.0801	0 4368	0.83	0.84±0.007
4	0.019	0 398	0.380	1.1215	0 4535	0 84	
1	0.011	0 456	0 445	0 7831	0.3115	1.43	
2	0.018	0.475	0.457	0 7769	0.3091	1.48	
3	0.019	0.463	0.444	0 7815	0 3109	1.43	1.45±0.021
4	0 019	0 471	0.452	0.7915	0.3149	1.44	
1	0 014	0.118	0.104	1.1919	0.4751	0.22	
2	0.022	0 124	0 102	0.9090	0 3623	0.28	
3	0 021	0 133	0 112	1.1545	0.4602	0.24	0.23±0.033
4	0.011	0 110	0 099	1.2991	0.5178	0 19	
1	0 022	0 155	0 133	0 6954	0 2789	0.48	
2	0.011	0.152	0.141	0 8274	0.3318	0.42	
3	0.016	0 154	0.138	0.6943	0 2784	0.50	0.47±0.03
4	0 019	0.156	0.137	0.7070	0 2835	0.48	



PROTEIN EFFECIENCY RATIO
1 4
1 3
1 2
1 1
1 0
0 9
0 8
0 7
0 6
0 5
0 4
0 3
0 2
0 1
FM
SQM
CM
PHM
SYM
FEEDS

Table 10. Protein digestibility co-efficient of various formulated diets fed to <u>C.chanos</u> fry

Replication	Average amount of food consumed (g)	Average amount of excreta produced (g)	Protein in feed %	Protein in excreta %	Protein in food consumed a (g)	Protein lost through excreta b (g)	Protein digested a-b=c (g)	Protein digestibility of co-efficient c/a	Mean ± SD
1	0.487	0.128			0.196	0.0267	0.1693	36.37	
2	0.510	0.131			0.205	0.0278	0.1777	86.68	
3	0.513	0.130	40.28	20.88	0.207	0.0271	0.1799	86.90	86.565±0.239
4	0.486	0.128			0.195	0.0267	0.1683	86.31	
1	0.512	0.080			0.209	0.0170	0.1920	91.87	
2	0.523	0.082			0.212	0.0171	0.1949	91.93	
3	0.514	0.084	40.44	20.83	0.208	0.0175	0.1905	91.59	91.86±0.172
4	0.529	0.082			0.214	0.0170	0.1970	92.06	
1	0.373	0.019			0 148	0.0033	0.1447	97.77	
2	0.369	0.018			0.147	0.0032	0.1438	97.82	
3	0.370	0.027	39.78	17.61	0.147	0.0049	0.1423	96.67	97.24±0.558
4	0.372	0.028			0.148	0.0049	0.1431	96.69	
1	0.592	0.323			0.236	0.0810	0.1550	65.63	
2	0.433	0.239			0.173	0.0600	0.1330	65.31	
3	0.549	0.304	39.86	25.00	0.219	0.0760	0.1430	65.30	65.23±0.379
4	0.618	0.338			0.246	0.0870	0.1590	64.63	
1	0.331	0.130			0.133	0.0294	0.1036	77.89	
2	0.394	0.155			0.158	0.0352	0.1228	77.72	
3	0.330	0.130	40.10	22.68	0.132	0 0295	0.1025	77.65	77.78±0.097
4	0.336	0.132			0.135	0 0299	0.1051	77.85	

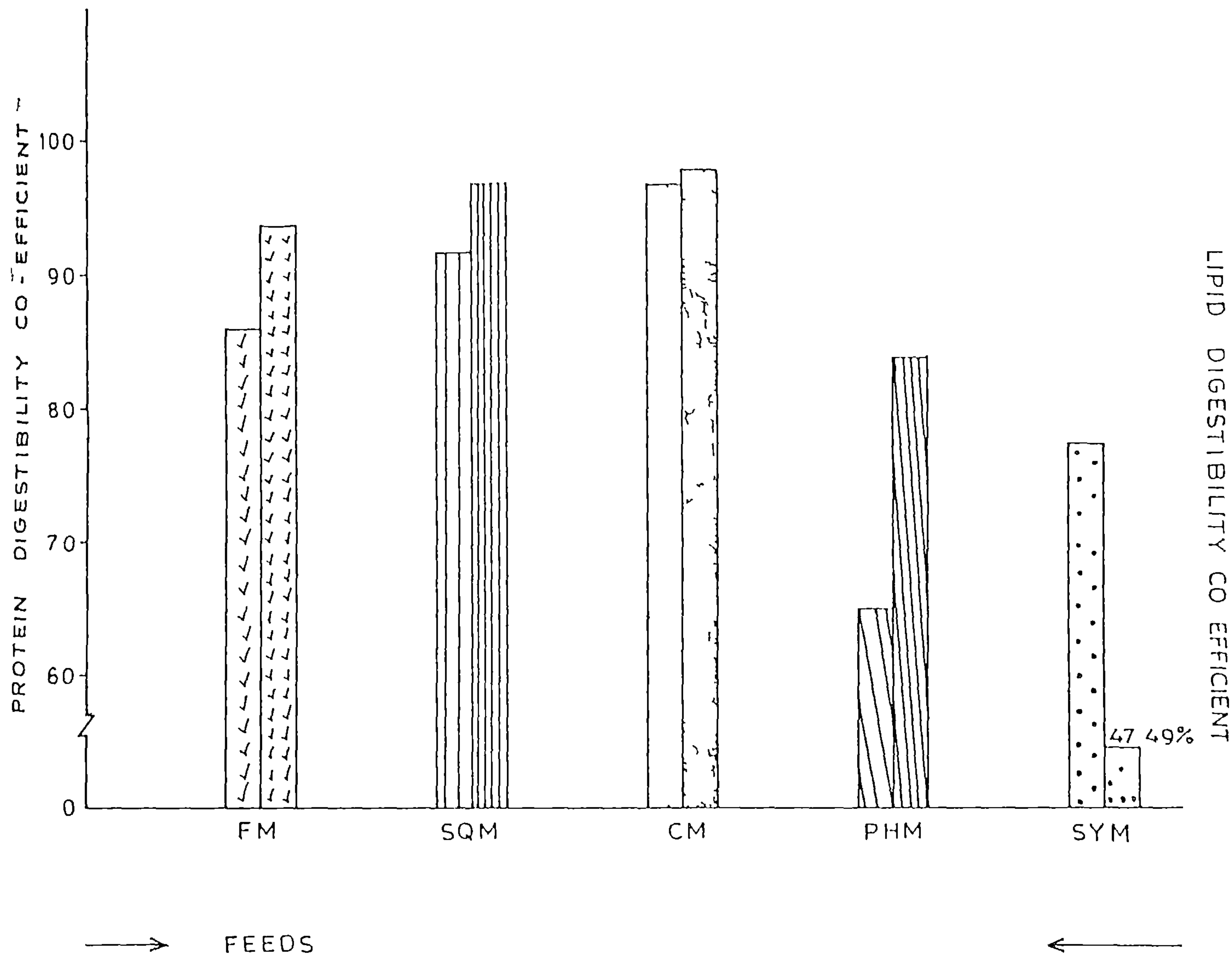

PROTEIN DIGESTIBILITY CO-EFFICIENT
100
90
80
70
60
0
FM
SQM
CM
PHM
SYM
47.49%
LIPID DIGESTIBILITY CO EFFICIENT
FEEDS

Table 11. Lipid digestibility co-effacient of various formulated diets fed to <u>C. chanos</u> fry.

Feed	Replication	Average amount of food consumed g	Average amount of excreta produced g	Lipid in feed %	Lipid in excreta %	Lipid in food consumed a g	Lipid lost through excreta b g	Lipid digested a-b=c g	Lipid digestibility co-effecient c/a	Mean ± SD
	1	0 487	0 128			0 032	0 0020	0 0300	93 75	
	2	0 510	0 131			0 033	0 0020	0 0310	93 94	
FM	3	0 513	0 130	6 5	1 6	0 033	0 0020	0 0310	93 94	93 845±0 09
	4	0 483	0 128			0 032	0 0020	0 0300	93 75	
	1	0 512	0 080			0 036	0 0004	0 0356	98 89	
	2	0 523	0 082			0 037	0 0004	0 0357	96 49	
SCM	3	0 514	0 084	7 1	0 46	0 036	0 0004	0 0356	98 89	97 12±1 94
	4	0 529	0 082			0 038	0 0004	0 0358	94 21	
	1	0 373	0 019			0 030	0 0005	0 0295	98 30	
	2	0 369	0 018			0 030	0 0004	0 0296	98 67	
CM	3	0 370	0 027	8 1	2 46	0 030	0 0007	0 0293	98.67	98 08±0 43
	4	0 372	0 028			0 030	0 0007	0 0243	97 67	
	1	0 592	0 323			0 043	0 0070	0 0360	83 72	
	2	0 433	0 239			0 031	0 0050	0 0260	83 87	
PHM	3	0 549	0 304	7 2	2 1	0 040	0 0060	0 0340	85 00	84 17±0 497
	4	0 618	0 338			0 044	0 0070	0 0370	84 09	
	1	0.331	0 130			0 019	0 0100	0 0090	47 37	
	2	0 394	0 155			0 023	0 0120	0 0110	47 83	
SYM	3	0 330	0 130	5 8	7 6	0 019	0 0100	0 0090	47 37	47 49±0 199
	4	0 336	0 132			0 019	0 0100	0 0090	47 37	

significantly different from CM (Table 11) The values for the digestibility of other feeds are given in Table 11 Lipid from SYM was the least digestible (47 49 + 0 19%) for C chanos fry (Table 11, Fig 4) Hence it seems that lipid from animal sources is more digestible than from plant source

4.5.3 Biochemical composition of C.chanos fry.

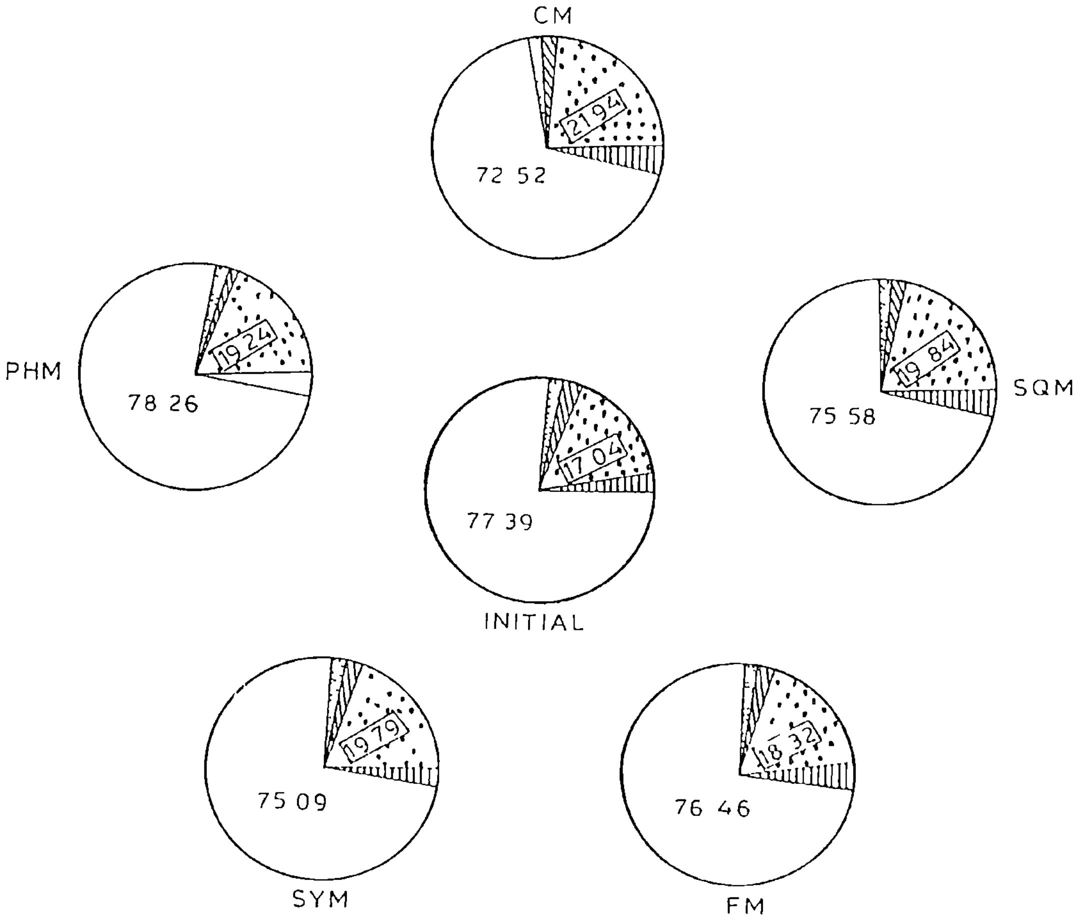
The biochemical composition of fish fed on various supplementary feeds is given in Table 12 Moisture content of fish decreased slightly (range being -0 3 to -4 87%), when fed with FM, SQM, CM or SYM However, fish fed with PHM showed an increase (0 87%) in moisture content The maximum increase in fish body protein was found when fed with CM, the increase being +4 9% This was followed by SQM (+2 8%) and FM (+1 10%) The above data also reflects the same order of performance in terms of PLP and protein digestibility co-efficient of various feeds when fed to C chanos fry Protein synthesis was low (0 75% increase) in fish fed on SYM, while it was the lowest (0 2%) with PHM (Fig 5) However, crude fat showed a different pattern of deposition It was maximum in fish fed on CM (0 89% increase) and minimum in fish fed on plant source SYM (0 15% increase) Fish fed on PHM produced better fat deposition (+0 28%) than those fed with FM (+0 20%). Fat deposition in fish fed on SQM showed an increase of 0 38% Maximum ash deposition was found in fish fed on PHM, while the lowest was found in those fed on SQM The range of percentage increase in ash deposition was in the order of +0 08 to +0 35% The NFE content in general showed a decrease in the fish body after feeding with

Table 12. Biochemical composition of C. chanos fry fed with various formulated feeds *

Parameter % basis	Initial	Feeds				
		FM, % difference	SQM, % difference	CM, % difference	PHM, % difference	SYM % difference
Moisture	77 39	76 64, (-0 75)	75 58, (-1 81)	72 52, (-4 87)	78 26, (+0.87)	77 09, (-0 3)
Crude protein **	17 04	18 14, (+1 10)	19 84, (+2 8)	21 94, (+4 9)	17 24, (+0 2)	17 79, (+0.75)
Crude fat	1 01	1 21, (+0 20)	1 39, (+0 38)	1 9, (+0 89)	1 29, (+0 28)	1 16, (+0 15)
Ash	1 55	1.71, (+0 16)	1 63, (+0 08)	1 69, (+0 14)	1 90, (+0 35)	1 69, (+0.14)
NFE	3 01	2 30, (-0.71)	1 56, (-1 45)	1 95, (-1 06)	1 31, (-1 7)	2 27, (-0 74)
Energy content Kcal/100 g	95 80	100 00, (+4 2)	106.60, (+10 8)	121 70, (+25 9)	93 13, (-2 67)	97 0, (+2 1)

Average four values

Expressed on wet weight basis


CM
21 94
72 52
PHM
19 24
78 26
SQM
19 84
75 58
17 04
77 39
INITIAL
19 79
75 09
SYM
18 32
76 46
FM

various supplementary feeds The maximum reduction in NFE was found in fish fed on FM (-0 71%). Energy content showed an increase in its value after fish were fed with various feeds, except PHM feed, the maximum being in those fed on CM (+25.9%) However, the fish fed on PHM showed a decrease (-2.67%) in the energy content.

4.5.4 Oxygen consumption, ammonia excretion and O:N ratio.

The Oxygen consumption of C chanos fry varied from 0 48 to 0 665 mg O_2 /g/hr, when fed with various feeds compounded from different protein sources. In general, fish fed with animal protein sources consumed more oxygen than that fed on plant protein source, except PHM (Table 13). The statistical analysis showed that oxygen consumption rate was not significantly ($P > 0$ 01) different among fish fed SQM and FM, and CM and SYM (Table 14).

Ammonia excretion rates of C chanos fry ranged from 0 043 to 0.117 mg NH_3 -N/g/hr (Table 13) when fed with different protein sources. Its value was lowest with fish fed on animal protein sources (except PHM-0 117 mg NH_3 N/g/hr). Fish fed on SYM showed a value of 0.091 mg NH_3 - N/g/hr Analysis of Variance (Table 14) shows no significant difference ($P \leq 0$ 05) of ammonia excretion between fish fed on PHM, SYM and FM or between SYM, FM and SQM or between FM, SQM and CM.

O N ratio was 12 566 in fish fed on CM, 10 769 for SQM, 8 388 for FM, 6 061 for SYM and 4 09 for PHM The ratio was low with fish

Table 13. Oxygen consumption, Ammonia excretion and O:N ratio in *C.chanos* fry fed on formulated feeds compounded from various protein sources

Parameters *	Feeds (Mean ± SD)				
	FM	SQM	CM	PHM	SYM
Length (cms)	3 361±0 010	3.819±0.0930	4 305±0.061	2.13±0 095	2 703±0.1010
Weight (g)	0 307±0.081	0.385±0.0730	0.468±0.015	0.121±0.011	0.153±0.0890
Oxygen consumption rate (metabolic rate) mgO_2/g/hr	0 651±0.003	0.665±0 0170	0.5405±0.024	0.480±0 026	0.5515±0 0180
Ammonia excretion rate (NH_3N/g/hr)	0 073±0 003	0.0618±0.0015	0 043±0 001	0 117±0 002	0.091+0.00090
O N ratio	8.388±0 400	10 769±0 0840	12 566±0 260	4 098±0 220	6.061±0 1980

* Average of four values.

Table 14. ANOVA Table for efficiency of formulated feeds fed to C.chanos fry.

Parameters	Source	Degrees of freedom	Sum of Squares	Mean Sum of Squares	F Value	N/S at 1%
FCR	Between feeds	4	318 008	79.522	135.17	S
	Error	15	8.825	0.5883		
	Total	19	326.913			
PER	Between feeds	4	38 403	9 6008	592.462	S
	Error	15	0.243	0.0162		
	Total	19	38.403			
Protein digestibility coefficient	Between feeds	4	1706.372	426.59	1628.21	S
	Error	15	3.932	0.262		
	Total	19	1710.304			
Lipid digestibility coefficient	Between feeds	4	3996.005	999.001	261.12	S
	Error	15	57.382	3.825		
	Total	19	4053 387			
Oxygen consumption	Between feeds	4	1.412	0.353	44.175	S
	Error	15	0.119	0.008		
	Total	19	1.531			
NH_3 excretion	Between feeds	4	1.384	0.346	4.669	S*
	Error	15	0 111	0.0741		
	Total	19	2.496			
O N ratio	Between feeds	4	214.54	53.635	505.99	S
	Error	15	1.59	0.106		
	Total	19	216.130			

N - not Significant, S - Significant

S* Significant at 5% level.

Comparison of treatment means based on critical difference

FCR

Standard error of transformed treatment means = 0.542

Critical difference = 1.598

Diets	PHM	SYM	FM	SQM	CM
Transformed treatment means	19.280	13.333	11.013	9.873	7.589

(FM and SQM are underlined together.)

PER

Standard error of transformed treatment means = 0.09

Critical difference = 0.265

Diets	CM	SQM	FM	SYM	PHM
Transformed treatment means	6.904	5.259	4.739	3.929	2.757

Protein Digestibility co-efficient

Standard error of transformed treatment means = 0.381

Critical difference - 1.1228

Diets	CM	SQM	FM	SYM	PHM
Transformed treatment means	80.481	73.426	68.499	61.875	53.867

Lipid Digestibility co-efficient

Standard error of transformed treatment means = 1.383

Critical difference = 4.075

Diets	CM	SQM	FM	PHM	SYM
Trnasformed treatment means	82.181	80.796	74.886	66.557	43.558

Oxygen consumption

Standard error of transformed treatment means = 0.063

Critical difference = 0.1864

Diets	SQM	FM	SYM	CM	PHM
Transformed treatment means	4.677	4.627	4.258	4.215	3.973

Ammonia excretion

Standard error of transformed treatment means = 0.192

Critical difference = 0.409

Diets	PHM	SYM	FM	SQM	CM
Transformed treatment means	1.962	1.729	1.597	1.424	1.183

<u>O N ratio</u>

Standard error of transformed treatment means = 0.230

Critical difference = 0.678

Diets	CM	SQM	FM	SYM	PHM
Transformed treatment means	20.757	19.157	16.83	14.25	11.674

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* Underscored means are not significantly diffe ent at 1% level.

** Not significant at 5% level.

fed on animal protein sources, except for PHM O N ratio of fish fed on different feeds was significantly different (Table 14)

4.5.5 Performance of various Feeds based on Survival and Growth of C.chanos fry.

4.5.5.1 Based on rate of Survival.

The rate of survival of C chanos fed with various formulated feeds is given in Table 15 The lowest survival was found in fish fed on PHM (81.25 ± 18 75%) and the highest in fish fed on CM or SQM (96.88±5.41%). Statistical analysis shows no significant difference (P > 0 01) between the rate of survival among fish fed on different feeds (Table 18)

4.5.5.2 Based on Growth

There was a clear-cut difference in growth in C chanos fry fed with different formulated feeds (Plate III) The highest average gain in length and weight was attained by fish fed on CM, the values being 2.55 cms and 0 450 g respectively (Table 16) The least gain in growth (0.6 cm in length and 0 104 g in weight) was obtained for fish fed on PHM (Fig. 6) The gain in length and weight of fish fed on FM (1 8 cms and 0.288 g respectively) was even significantly lower (P ≤ 0.01) than those fed on SQM (2 175 cms and 0 368 g respectively) The gain in growth was 1 025 cms and 0 137 g when plant source of protein (SYM) alone was used (Table 16)

Table 15. Rate of survival of <u>C.chanos</u> fry fed on different formulated diets (on % basis)

Treatment	Replications				
	1	2	3	4	5
FM	87.5	100.0	100.0	75.0	90.63±10.36
SQM	87.5	100.0	100.0	100.0	96.88± 5.41
CM	100.0	87.5	100.0	100.0	96.88± 5.41
PHM	87.5	50.0	87.5	100.0	81.25±18.75
SYM	87.5	100.0	87.5	87.5	90.63± 5.41

Plate III

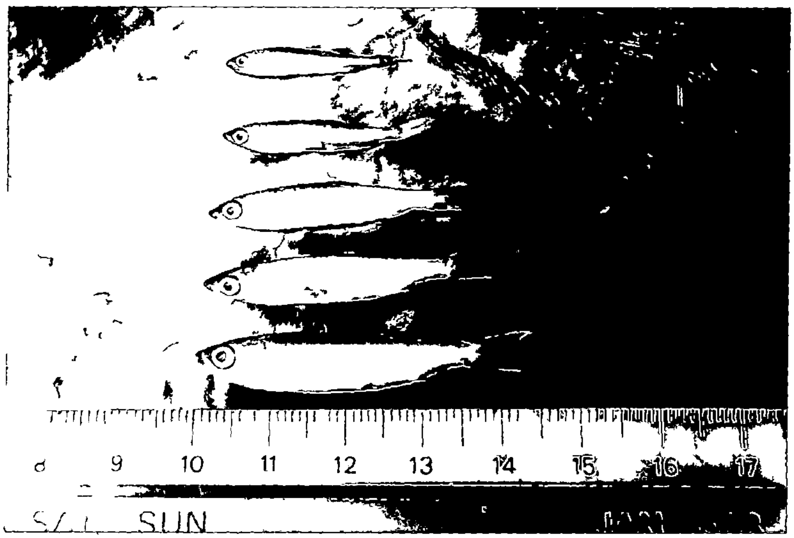
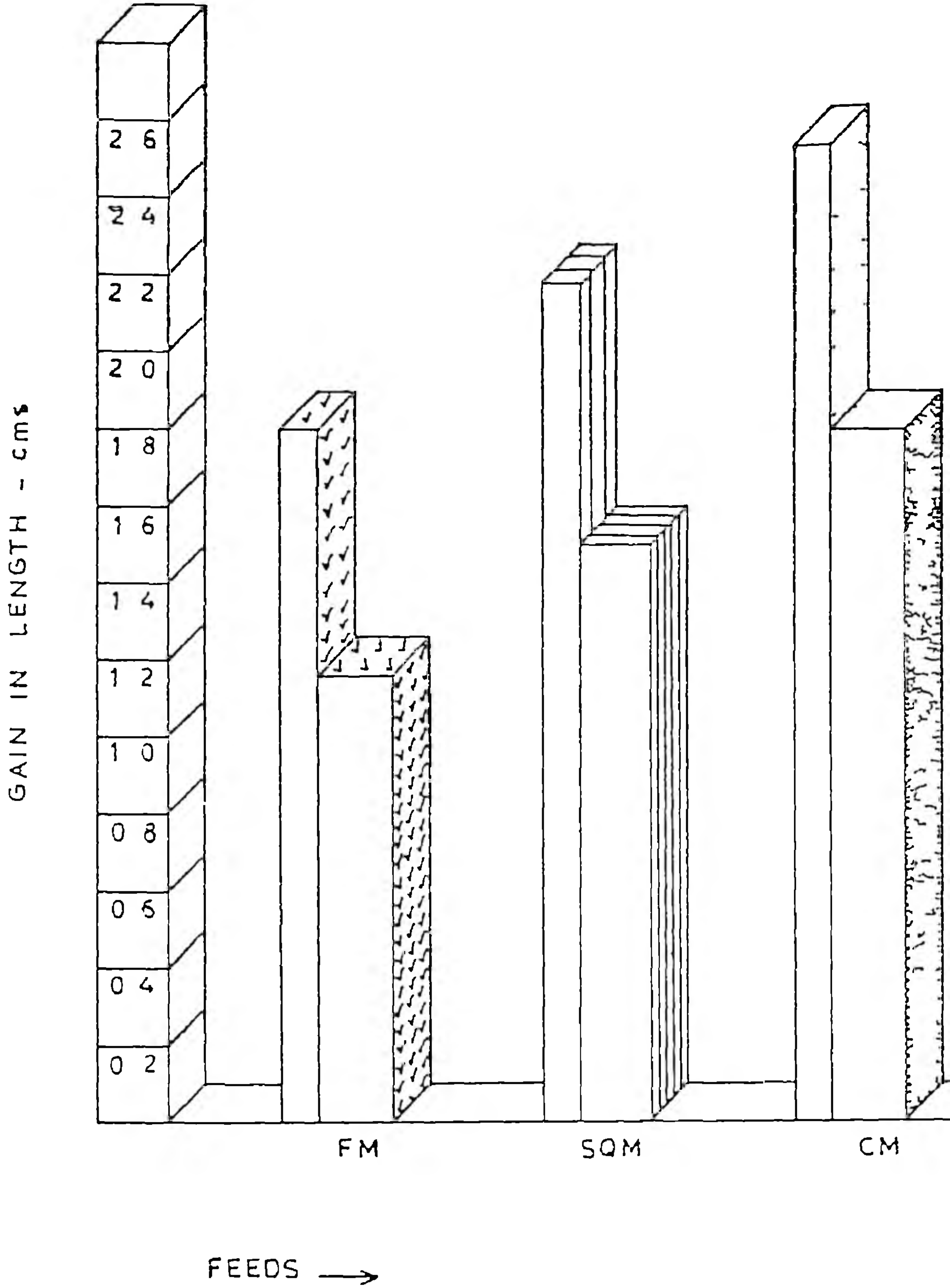
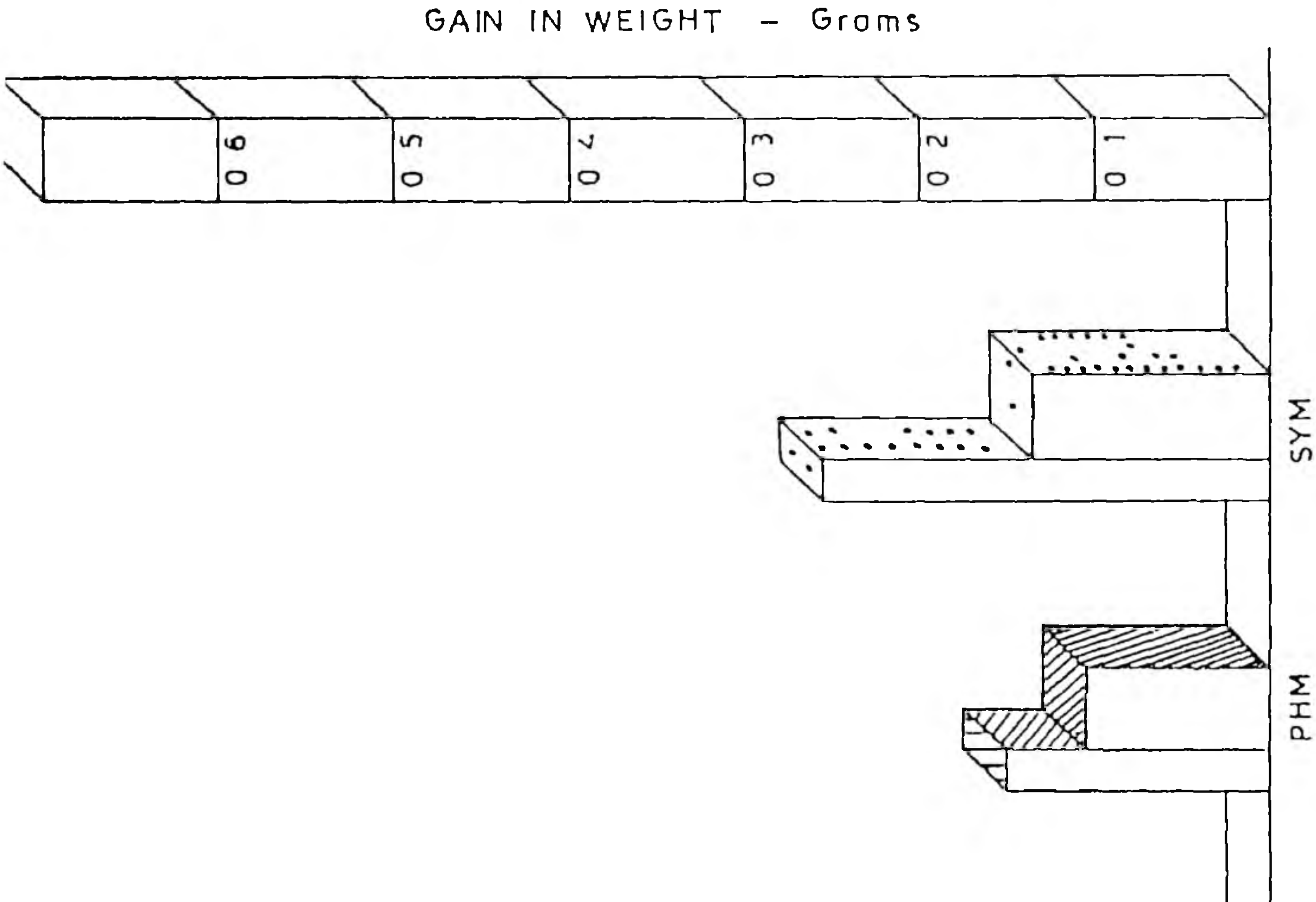


Table 16. Average length (cm) and weight (g) gain, and percentage increase in growth attained by C.chanos fry fed with various diets for 42 days (Values in paranthesis give the average)

Diet	Replication	Initial		Final		Net gain		Growth (%)	
		Length cm	Weight gm	Length cm	Weight gm	Length cm	Weight gm	Length	Weight
FM	1	1.7	0.019	3.7	0.312	2.0	0.293	117.65	1542.11
	2	1.6	0.011	2.6	0.303	2.0	0.292	125.00	2654.55
	3	1.9	0.022	3.6	0.307	1.7	0.285	89.47	1295.45
	4.	2.1	0.028	3.6	0.307	1.5	0.279	71.43	996.43
		(1.825)	(1.020)	(3.375)	(0.307)	(1.800)	(0.288)	(100.890)	(1622.140)
SQM	1	1.6	0.011	3.8	0.377	2.2	0.366	137.5	3327.27
	2	1.7	0.015	3.9	0.394	2.2	0.379	129.41	2526.67
	3	1.7	0.011	3.9	0.375	2.2	0.364	129.41	3309.09
	4	1.9	0.019	4.0	0.398	2.1	0.380	105.26	2000.00
		(1.725)	(0.014)	(3.900)	(0.385)	(2.175)	(0.368)	(125.400)	(2790.76)
CM	1	1.6	0.011	4.2	0.456	2.6	0.445	162.5	4045.45
	2	1.9	0.018	4.4	0.475	2.5	0.457	131.58	2538.89
	3	1.8	0.019	4.3	0.463	2.5	0.444	138.89	2336.84
	4.	1.8	0.019	4.4	0.471	2.6	0.452	144.44	2378.95
		(1.775)	(0.017)	(4.325)	(0.466)	(2.550)	(0.450)	(144.270)	(2825.030)
PHM	1	1.8	0.014	2.4	0.118	0.6	0.104	33.33	742.86
	2	1.9	0.022	2.3	0.124	0.4	0.102	21.05	463.64
	3	1.7	0.021	2.6	0.133	0.9	0.112	52.94	533.33
	4	1.9	0.011	2.4	0.110	0.5	0.099	26.32	900.00
		(1.825)	(0.017)	(2.425)	(0.121)	(0.600)	(0.104)	(33.410)	(659.960)
SYM	1	2.0	0.022	3.0	0.155	1.0	0.133	50.00	604.55
	2.	1.6	0.011	2.7	0.152	1.1	0.141	68.75	1281.82
	3.	1.7	0.016	2.8	0.154	1.1	0.138	64.71	862.5
	4.	1.9	0.019	2.8	0.156	0.9	0.137	47.37	721.05
		(1.800)	(0.017)	(2.825)	(0.154)	(1.025)	(0.137)	(57.710)	(867.48)



GAIN IN LENGTH - cms
2 6
2 4
2 2
2 0
1 8
1 6
1 4
1 2
1 0
0 8
0 6
0 4
0 2
FM
SQM
CM
FEEDS


GAIN IN WEIGHT - Grams
0 1
0 2
0 3
0 4
0 5
0 6
SYM
PHM

Analysis of Variance indicates that gain in length and weight were significantly ($P \leqslant 0.01$) influenced by the different protein sources used in the feed (Table 18). Maximum gain in growth was obtained in diets compounded with animal sources of protein except for prawnhead waste meal.

Table 16 also gives the percentage gain in growth of C chanos fry fed on different feeds, which again reflects the same order of performance of feeds. The percentage increase in length of fish was 144.27, 125.4, 100.89, 57.71 and 33.41%, and the increase in weight, 2825.03, 2790.76, 1622.14, 876.48 and 659 96% for CM, SQM, FM, SYM and PHM respectively.

4.5.5.3 Based on Specific Growth Rate.

The SGR was the highest for fish fed on CM (7 98 ± 0.52) and the lowest for PHM (4.78 ± 0.54) (Fig.2) The SGR values for other feeds are given in Table 17

Statistical analysis showed that SGR of fish fed on CM and SQM was significantly higher ($P \leqslant 0.01$) than the others However the values obtained for fish fed on plant source only (SYM) was not significantly different from FM, the values for SYM and PHM were also not significantly different from each other

Table 17. Specific growth rate of C. chanos fry fed with various diets for 42 days.

Feed	Replication	Average initial weight (g)	Average final weight (g)	Specific growth rate SGR (%)	Mean ± SD
	1	0.019	0.312	6.66	
FM	2	0.011	0.303	7 89	6.63±0 80
	3	0.022	0.307	6.28	
	4	0 028	0.307	5.70	
	1	0 011	0.377	8.42	
SQM	2	0 015	0.394	7.78	
	3	0 011	0.375	8.40	7.96±0.49
	4	0.019	0.398	7.24	
	1	0 011	0 456	8 87	
CM	2	0 018	0.475	7 79	
	3	0 019	0.463	7 60	7.98±0.52
	4	0 019	0.471	7.64	
	1	0.014	0.118	5 08	
PHM	2	0 022	0.124	4.12	
	3	0 021	0.133	4.39	4.78±0.54
	4	0.011	0.110	5 48	
	1	0.022	0.155	4.65	
SYM	2	0.011	0.152	6.25	
	3	0.016	0.154	5.39	5.33±0.59
	4	0.019	0.156	5.01	

Table 18. ANOVA Table for the performance of feed on the survival and growth of *C. chanos* fry.

Parameters	Source	df	SS	MSS	F value	N/s at F 0.01%
Rate of Survival	Between feeds Error Total	4 15 19	792.958 2666.238 3459.196	198.240 177.749	1.115	N
Gain in length	Between feeds Error Total	4 15 19	10.397 0.365 10.762	2.599 0.024	106.8333	S
Gain in weight	Between feeds Error Total	4 15 19	0.352 0.00058 0.35283	0.008 0.000039	2257.95	S
SGR	Between feeds Error Total	4 15 19	48.3101 9.9558 58.2659	12.078 0.66372	18.197	S

N- Not significant, S - significant.

Comparison of treatment means based on critical difference.

Gain length

Standard error of treatment means = 0.1103

Critical difference = 0.325

Diets	CM	SQM	FM	SYM	PHM
Treatment means	2.55	2.175	2.55	1.025	0.6

Gain in weight

Standard error of treatment means - 0 0044

Critical difference = 0 013

Diets	CM	SQM	FM	SYM	PHM
Treatment mean values	- 2 55	0 372	0 287	0 137	0 104

SGR

Standard error of transformed treatment means - 0 576

Critical difference = 1 698

Diets	CM	SQM	FM	SYM	PHM
Transformed treatment means	16 396	16 380	14 499	13 322	12 396

* Underscored means are not significantly different at 1% level

4.6 Studies on Optimum Food Ration

4.6.1 Water quality

Water quality parameters recorded during the study for optimum food ration is given in the Table 19 Salinity of water was maintained at 4 to 6 ppt The water temperature ranged from 28 6 to 29 3°C pH fluctuated between 8 2 to 8 6, and the dissolved oxygen from 5 9 to 8 1 ppm.

4.6.2 Rate of Survival.

Survival rate was the poorest (66 67 ± 9 43) at zero level of feeding It was only 70 ± 8 16 at 18% level of feeding At 3% level of feeding the survival rate was 80% and at 6% level it was 100% At all other levels, the rate of survival was between 80 and 100% (Table 20)

The statistical analysis (Table 21) shows that survival was not significantly different ($P > 0\ 01$) at 3%, 6% or 12% level of feeding

4.6.3 Specific Growth Rate.

SGR attained a negative value of -1 14 at zero level of feeding It was 2 02 at 3% and 4 67 at 6% level of feeding Specific growth rate increased from -1.14 to 7 63 and then decreased to 6 37 when fed at 0, 12 and 21% of body weight, respectively (Fig 7, Table 20)

Table 19. Water quality parameters of the experimental tanks during the study for optimum food ration in *C. chanos* fry.

Parameters	Range	Mean + SD
Water temperature in °C	28.6 - 29 3	28 73 ± 0 45
Salinity in ppt	4 - 6	5 + 0 05
pH	8 2 - 8.6	8.25 + 0.12
Dissolved oxygen in ppm	5.9 - 8 1	6 98 + 0 34

Table 20. Effect of food ration on survival, growth and food conversion ratio in *C.chanos* fry.

Feeding rate	Replication	Initial weight g	Final weight g	Gain in weight g	SGR %	Mean ± SD SGR %	Food consumed g	FCR	Mean±SD FCR	Rate of survival	Mean ± SD Rate of survival
	1	0.114	0.089	-0.025	-1.20		0.000	-	-	80	66.67±9.43
0	2	0.116	0.093	-0.023	-1.05	-1.14±0.07	0.000			60	
	3	0.118	0.092	-0.025	-1.18		0.000			60	
	1	0.106	0.162	0.056	2.01		0.101	1.80		80	
3	2	0.100	0.152	0.052	1.99	2.02±0.029	0.098	1.88	1.83±0.04	80	80±0.00
	3	0.100	0.154	0.054	2.06		0.098	1.81		80	
	1	0.105	0.281	0.176	4.68		0.333	1.90		100	
6	2	0.101	0.279	0.178	4.83	4.66±0.15	0.330	1.89	1.91±0.03	100	100±0.00
	3	0.112	0.286	0.174	4.46		0.340	1.95		100	
	1	0.110	0.439	0.329	6.59		0.663	2.02		80	
9	2	0.106	0.433	0.331	6.70	6.58±0.10	0.666	2.01	2.01±0.005	80	80±0.00
	3	0.114	0.433	0.329	6.46		0.661	2.01		80	
	1	0.100	0.498	0.392	7.64		0.852	2.17		80	
12	2	0.101	0.501	0.400	7.63	7.63±0.01	0.852	2.13	2.15±0.02	100	93.3±9.43
	3	0.100	0.496	0.396	7.62		0.850	2.15		100	
	1	0.106	0.491	0.385	7.30		0.942	2.45		80	
15	2	0.103	0.486	0.383	7.39	7.38±0.07	0.942	2.46	2.45±0.01	100	93.3±9.43
	3	0.102	0.489	0.387	7.46		0.941	2.43		100	
	1	0.102	0.463	0.361	7.20		0.990	2.74		60	
18	2	0.101	0.462	0.361	7.24	7.18±0.06	0.992	2.75	2.75±0.01	70	70±8.16
	3	0.105	0.465	0.361	7.09		1.000	2.77		70	
	1	0.113	0.430	0.317	6.36		0.998	3.15		100	
21	2	0.113	0.432	0.319	6.38	6.37±0.01	0.998	3.13	3.15±0.01	100	100±0.00
	3	0.113	0.427	0.315	6.37		0.998	3.17		100	

Analysis of Variance (Table 21) shows significantly ($P \leq 0.01$) lower SGR when fed at low levels of feeding

4.6.4 Food Conversion Ratio.

FCR was best at low levels of feeding, it increased from 1.83 to 3.15 when fed from 3% to 21% of body weight

Analysis of Variance shows that FCR is significantly different ($P \leq 0.01$) at all levels of feeding (Table 21)

4.6.5 Extrapolation of Optimum Food Ration from graph.

The extrapolation of graph (Fig 7) between SGR and feeding rate indicate that the optimum food ration of C chanos fry is when fed at a level of 5.2% of the body weight The maintenance ration is at 1% of the body weight and the maximum growth rate at 12% of the body weight.

Table 21. ANOVA Table optimum food ration of C.chanos fry.

Parameters	Source	df	SS	MSS	F	N/s at F(7.16)
Rate of survival						
	Between feeding rates	7	4070.77	581.475	8.63	S
	Error	16	1077.084	67.318		
	Total	23	5147.8538			
Specific Growth Rate						
	Between feeding rates	7	404.975	57.854	732.329	S
	Error	16	1.263	0.079		
	Total	23	406.238			
Food Conversion Ratio						
	Between feeding rates	7	69.511	9.93	6895.83	
	Error	16	0.023	0.00144		S
	Total	23	69.534			

N - not significant at 1% level.

S - significant at 1% level.

Comparison of treatment means based on critical difference.

Rate of survival

Standard Error of transformed treatment means = 6.699

Critical difference = 19.568

Feeding rates	6	21	12	15	3	9	18	0
Transformed treatment means	90	90	81.43	81.43	63.43	63.43	61.22	54 99

Specific Growth Rate

Standard Error of transformed treatment means = 0 229

Critical difference = 0 670

Feeding rate	12	15	18	9	21	6	3	0
Transformed treatment means	18 077	17 838	17 633	17 035	16 817	14.951	11 978	5 306

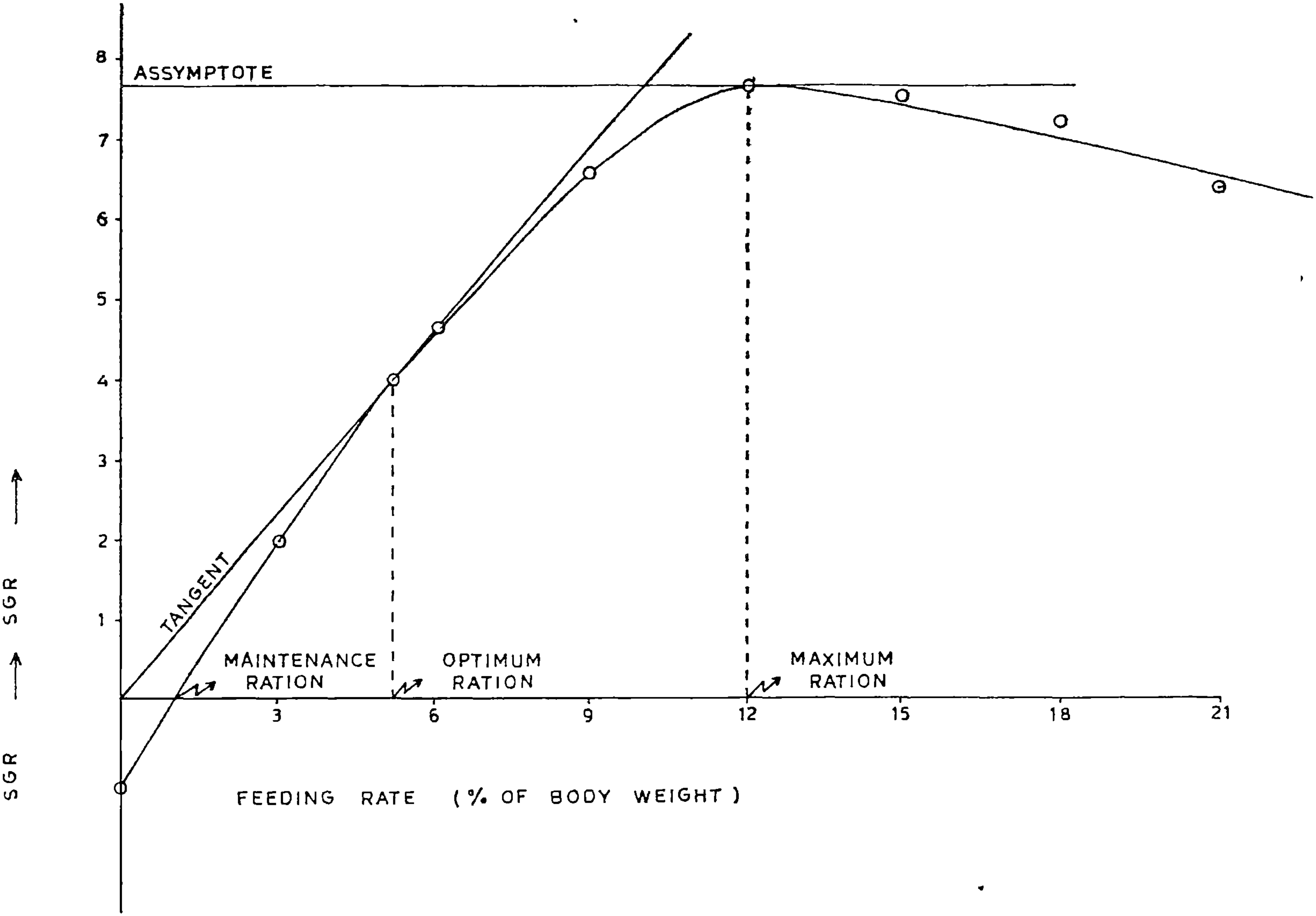
FCR

Standard Error of transformed treatment means = 0 001144

Critical difference = 0.0905

Feeding rate	21	18	15	12	9	6	3	0
Transformed treatment means	35 263	33 513	32 097	30 669	29 99	29 483	29 054	17 217

* underscored means are not significant at 1% level


ASSYMPTOTE
TANGENT
MAINTENANCE RATION
OPTIMUM RATION
MAXIMUM RATION
SGR
SGR
FEEDING RATE (% OF BODY WEIGHT)
1
2
3
4
5
6
7
8
3
6
9
12
15
18
21

5 DISCUSSION

5.1 Keeping Quality of Formulated Feeds

A good quality feed is one which is able to maintain its nutritional status for a few weeks of storage. According to Fowler and Banks (1967) nutritional status of feed does not show any alteration if stored for a few weeks, but if stored for a longer period, it may have deleterious effects on fish growth. The shelf-life of the processed feeds is dependent on the type of processing, storage temperature and moisture content of the diet (Hilton et al., 1977) During prolonged storage adverse physical conditions (moisture, heat, light) and micro-organisms (mould, yeast and fungi) can cause deterioration of feed quality resulting in decreased palatability and nutritive value (Chow, 1980). According to Hardy (1989) dry feeds should be used within 90 days of production. However, feeds used in the present study did not show any drastic variation in the nutritional status, even after four months of storage, indicating that the quality of feeds developed were satisfactory.

5.1.1 Moisture.

There was an increase in the moisture content of all feeds after four months of storage. Hastings (1971), reports that the presence of hygroscopic ingredients mainly starch brings about softening of the pellets during storage. This could have resulted in the maximum absorption of moisture by SYM - the feed which had fairly high amount

DISCUSSION

of carbohydrate content. Besides, SYM had the least fat content among the formulated feeds. Jayaram and Shetty (1981) report that higher fat content hardens the particles thereby preventing the entry of moisture into the pellets. This is also reflected in the lower moisture absorption of CM, SQM and FM which had higher fat content than SYM. The physical structure of feed ingredients and the porosity of feeds also affect the uptake of moisture (Srikanth, 1986). Since SYM absorbed more moisture it may be assumed that it was more porous. The higher crude fibre content of SYM could also have resulted in higher moisture absorption, as is also reported by Srikanth (1986) for the feeds he used.

The increase in the moisture content was accompanied by the increase in sinking rate. Hence SYM also recorded the maximum increase in the sinking rate. Similar observation has also been reported by Srikanth (1986) and Josekutty (1991).

5.1.2 Protein.

The slight decrease observed, in the protein level of all formulated feeds following storage may be due to their increased moisture content. Jayaram and Shetty (1981) attributed this to the breakdown of water-soluble proteins due to increase in moisture content, and Chow (1980) to the deterioration of amino acids. Borthakur (1983), Venugopal and Kesavanath (1984), and Josekutty (1991) have also observed similar conditions in the feeds they used.

5.1.3 Fat.

Among the five feeds tested, CM which had the highest fat content, also showed the highest reduction in the fat content after storage. Lipid content of the diet is seen to be inversely related to the moisture content. Jayaram and Shetty (1980 a), and Venugopal and Kesavanath (1984) attributed the decrease in fat content to an increase in the moisture content during storage and the degree of unsaturated oil present in the diet. Lipid auto-oxidation taking place during storage can also decrease the fat content. This may be one of the reasons for the reduced fat content after storage in the present study. However, there was no rancid smell for any of the feeds used in the present study even after four months of storage. The rate of oxidation of fat is greatly influenced by the storage temperature and a rise of 10°C approximately doubles the rate (Kulikov, 1978) De la Cruz et al. (1989) recommends that feeds should not be stored for more than 15 days during summer months when temperature ranges between 28 and 31°C. In the present study, temperature range was 27.1 to 30.8°C and the storage period four months. However, not much reduction in the fat content was observed. The order of decrease of fat content, after storage was CM > SQM > PHM > FM > SYM, which shows inverse relationship to the amount of moisture absorbed.

5.1.4 Ash, Crude fibre, Carbohydrate.

The reduction in ash and crude fibre content of the diet after storage may be attributed to the increase in the moisture content. The increase 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.

Total heterotrophic bacteria and fungi increased in all feeds after storage. This may be due to the absorption of moisture, during storage. Higher fat content can also attract the fungi which can deteriorate the feed. In the present study, CM which had the highest fat content showed maximum increase in total heterotrophic bacteria and fungi Cho et al. (1985) reported that the increase in microbial load resulted in the reduced palatability and nutritive value of feeds.

Oxidation of fat gives rise to a feedstuff of lower biological value (Rumsey, 1980). It can result in increased disease outbreaks like lipoid degeneration (Roberts, 1978). Oxidation can be reduced by lowering the storage temperature, controlling storage conditions (Kulikov, 1978) and by reducing oxygen levels during storage (Hardy, 1980). Natural antioxidants like tocopherol and chemical antioxidants like butylated hydroxy toluene (BHT) can be used to prevent lipid oxidation in feeds. Fungal growth can be prevented by the use of potassium metabisulphite, sorbic acid, benzoic acid or by propionic acid. Problems due to the chemical deterioration and infestation by microbes and insects can be reduced to some extent by storing the feed in air tight containers.

5.1.6 Water Stability.

The stability of feed is of utmost importance in aquaculture, as it makes the feed more efficient and economical. The stability of formulated feeds should be such that, it should remain stable in water atleast for one hour, so that they become available to even slow feeding fishes. The feed stability is influenced by ingredient composition, nature of ingredients, type of processing and by the moisture content of the diet (Hastings, 1971, Kainz, 1979).

The most stable feed both before and after storage was CM. This may be because of its low moisture absorption. High amount of glycogen in clam meal could also have resulted in the higher stability of CM feed. Hastings (1971) has stated that, higher fat content affect gelatinization, thereby reducing the stability of feed. Such a situation was not observed in the present study and the stability of feed was in the order of CM > SYM > SQM > FM > PHM. The least stable feed was PHM. Stivers (1970) and Jayaram and Shetty (1981), are of opinion that the degree of stability of feed is dependent on the gelatinization of starch during the cooking of feed. In the present study, the high amount of chitin and crude fibre in prawnhead waste might have resulted in poor gelatinization, affecting the stability of PHM. Such a situation has also been observed by Srikanth (1986).

However, too much stability of pellets is undesirable, since the nutrients get tied up and become unavailable to the organism (Balazs *et al*., 1973).

5.2 Water quality parameters in relation to Growth and Supplementary Feeding

C. chanos can tolerate wide ranges of temperature (8.5 - 42.7°C), salinity (0 - 158 ppt) and dissolved oxygen as low as 1.5 ppm (Chen, 1990). The fluctuations of salinity, temperature, pH, and dissolved oxygen in the present study were 4-6 ppt, 27.1 - 30.8°C, 7.6 - 8.2, 6.8 - 8.8 ppm respectively. Although, ammonia excretion rates were in the order of 0.043 - 0.117 mg NH_3 -N/g/hr, it was not reflected in the pH of water, since aeration was provided throughout the study. Hence the conditions that prevailed in the experimental tanks did not seem to affect the growth of fish.

5.3 Evaluation of Supplementary Feeds

5.3.1 Based on Efficiency of Feeds.

5.3.1.1 Food Conversion Ratio.

In the present study, better FCR values were recorded in fish fed with diets containing animal sources as chief protein, than that of plant source , except for PHM. The best FCR was given by CM (1.74), followed by SQM (2.94) and FM (3.65). The FCR of SYM was 5.32 which was however better than that of PHM (10.9).

Kiron (1989), in his studies of supplementary feeds in Liza parsia fry, obtained FCR of 1.8 for fish meal, 2.3 for soyabean meal and 2 65 for prawn waste based diets. Feeding trials conducted in ponds with *Cyprinus carpio*, using supplementary feed compounded from fish meal, silk worm pupae, clam meal and groundnut oil cake and rice bran have shown to yield an absolute conversion ratio of 3.94 to 5.3 (Borthakur, 1983).

The level of incorporation of various protein sources seems to influence the FCR. Seneriches and Chiu (1988) reported the betterment of FCR values from 4.9 to 1.5 as fish meal content in the diet was increased from zero to 30% of a corn meal based diet in *C. chanos* fry. Replacing FM from 0 to 40% of the SYM diet in *C. chanos* increased the performance of FCR values from 3.2 to 1.85 (Shiau *et al.*, 1988). A similar study in *Oreochromis mossambicus* fry (Davies *et al.*, 1989) indicated better performance of FCR from 2.6 to 2.19 when soyabean meal was reduced from 75% to 25% of the diet.

In *C. chanos* fry a formulated diet alone gave a better FCR (1.1), than a combination of formulated feed and *Spirulina* (1.5) or rice bran alone (1.9) (Santiago *et al.*, 1989). Among the different plant sources tested in *C. chanos* juveniles, soyabean meal gave a better FCR of 1.88 than mugo (2.05), pigeon pea (2.34) or kidney bean (3.31) (De la Pena *et al*, 1987).

The best FCR shown by clam meal diet in the present study, may be because of its high protein and lipid digestibility, indicating that CM protein is better utilized for growth purposes than other feeds.

5.3.1.2 Protein Efficiency Ratio.

Steffens (1981) has reported that PER values can be used to evaluate the quality of protein in the diet, those with higher PER are of good quality and those with lower PER values are of poor quality. Hence, in the present study, CM with the highest PER (1.45) can be considered as the best protein source among the five, followed by SQM (0.84) and FM (0.68). Soyabean is a poor protein source (0.47) while PHM is the poorest (0.23) for milkfish fry. A similar trend has also been observed in Liza parsia fry, by Kiron (1989) with fish meal, soyabean meal and prawn waste as protein sources.

5.3.1.3 Digestibility co-efficient.

Digestibility determination along with chemical analysis allow a more thorough estimation of nutritive value of a particular protein source (Plakas and Katayama, 1981). The digestibility co-efficient is found to vary with size of fish, microflora of the intestine and digestive enzymes (Nail, 1962 Shell, 1967, Syvokiene et al., 1975 Dabrowski, 1977). Digestibility may also be limited due to incomplete digestive action or to the incomplete absorption (Maynard and Loosli, 1978).

Protein digestibility co-efficient in the present study varies from 65.23 to 97.34%. Fish fed on CM and SQM showed values of 97.34 and 91.86% respectively. Borthakur (1983) has reported a slightly lower value (92.7%) for clam meal diet (included at 20% of the diet) in common carp. The protein digestibility of 86.56% obtained for FM in the present study compares well with the value of 86.16% obtained by Shiau et al. (1988) for C. chanos juveniles. However, the value was lower than those obtained for common carp (88.9 to 97.6%) Jayaram and Shetty 1980 b, c, Borthakur, 1983, Eid and Matty, 1987, Hossain and Jauncey, 1990) and for red tilapia (99 42%) (Kamarudin et al , 1989) Ogino and Chen (1973), attributed the wide variation in the protein digestibility of FM, to the origin and processing method used for the preparation of fish meal used in the study. In the present study, SYM had a protein digestibility of only 77.78% Although, similar results have been obtained for Nile tilapia (77.09%) by Wee and Shu (1989), the value is lower than that reported by Shiau et al (1989) for C. chanos (89.54%). Ferraris et al. (1986) have reported a still lower protein digestibility value of 54% for C. chanos fingerlings. These lower values of protein digestion of soyabean may be due to the trypsin inhibitors present in them (Smith, 1977, Dabrowski and Kozak, 1979), since only plant sources (soyabean meal and groundnut oil cake) were present in SYM feed. The protein digestibility values obtained for PHM (65.23%) in the present study, is much lower than that reported by Anil (1981) for carp (88.9%)& rohu (87.1%), and by Kiron (1989) for Liza parsia fry (76.34%). Meyers and Rutledge (1971) report that, although shrimphead waste contains good amount of digestible proteins rich in methionine and lysine, its utilization may be hampered by the high chitin or Ca salts present in it

The wide variation in protein digestion, inspite of equal protein levels in feeds, was attributed to the quality of protein type (Nose and Toyama, 1966, Nomura et al., 1971), biological availability of different proteins (Ogino and Chen, 1973) and to an unfavourable amino acid composition of the diet (Nose, 1963).

Lipid digestibility values, in general, were higher (84 to 98.08%) for fish fed with diets based on animal protein sources than with plant source (SYM - 47.49%). This may be because, the warmwater marine fishes have a higher requirement for n-3 HUFA (Rajyalakshmi, 1989). The feed sources used in the present study, viz , CM, SQM, PHM and FM are of marine origin and are probably rich in HUFA. SYM contained only plant sources which may contribute only to n-6 fatty acids. Bautista and de la Cruz (1988) have suggested linolenic acid (n-3) to be a better EFA for milkfish fingerlings than linoleic (n-6) acid, although growth and survival were not significantly different from each other. Besides, high amount of crude fibre in plant sources may lower the digestibility of nutrients because of rapid passage of food through the gut (Bender, 1967) Among the five feeds used in the present study, SYM had the highest crude fibre content (7.8%). These factors might have resulted in the poor lipid digestibility of SYM in milkfish fry.

The values of lipid digestibility obtained for CM (98.08%) and SQM (97.12%) are not significantly different Borthakur (1983) has reported lipid digestibility of 88.2% in common carp with clam meal diet (at 20% dietary inclusion). Lipid digestibility reported in the present study,

HUFA - Higher Unsaturated Fatty Acid.

for FM (93.8%) is found to be almost similar to those reported for common carp by Jayaram and Shetty (1980 b, c), Venugopal (1980), and Borthakur (1983), the values being 94.9, 96.5 and 90.4% respectively. Kiron (1989) has reported a lipid digestibility value of 91.85% for *Liza parsia* fry. However the present value of 84 17% for PHM, was lower than that reported for *Liza parsia* fry (92.18%) by Kiron (1989). Lipid digestibility of SYM (47.49%) used in the present study is much lower than that reported for *Liza parsia* fry (90.86%) by Kiron (1989).

In general, lipid digestibilities of feeds used in the present study, were higher than protein digestibility values, except for SYM. So it seems that lipid is the best source of energy for the fast swimming fishes like *C. chanos*. Moreover, Gorriceta (1982) and Gorriceta and Benitez (1983) have also detected lipase activity in almost all parts of the digestive tract of milkfish fry. This would also have contributed to the better utilization of lipid in *C. chanos* fry.

Besides being an energy source, lipids also act as feed attractor and increase the palatability of feed in fish and prawn (Rajyalakshmi, 1989). Chiu *et al* (1986 a) and Seneriches and Chiu, (1988) have observed schooling behaviour in *C. chanos* fry when fed with fish meal based diet, whereas, no such activity was observed with other diets. However, in the present study, no such differentiation was observed as schooling behaviour was exhibited by fry with all the diets offered. The schooling behaviour (See Plat IV) exhibited by fry as soon as the feed was given may be attributed to the natural feeding

Plate IV

stimutants reported to be present in squid meal, mussel meat and prawn head waste (Paulraj, 1989).

5.3.2 Based on biochemical composition of *C. chanos* fry before and after feeding.

The study of biochemical composition of fish is of paramount importance as the carcass composition allows the determination of optimum feeding rations. The growth of fish is influenced to a great extent by the chemical contents of the diet, age, and sexual maturity (Reimers and Meske, 1977, Viola and Amidan, 1978, Jayaram and Shetty, 1980 a, Reinitz and Hitzel, 1980, Zeitler *et al.*, 1984).

In the present study, a decrease in the moisture content and an increase in protein, fat, mineral and crude fibre contents were observed in fish fed on different diets. Tiews *et al.* (1973) and Huisman *et al.* (1979) reproted that in trials over a long period of time, a decline in the water content and an increase in fat content with minor changes in protein and mineral contents might be expected as weight increased with age in fish. In the present study, the moisture content of fish fed on various feeds ranged from 72.52 (CM) to 78.26% (PHM) It seems to have an inverse relationship with fat content of fish (except for fish fed on PHM), as reported by earlier investigators like Brandes and Dietrich (1958), and Love (1970). The increase in the moisture content of fish fed on PHM, inspite of the relatively high fat content in PHM may be due to the lower protein synthesis in these fishes.

The protein synthesis was in the order of CM > SQM > FM > SYM > PHM. A positive correlation of protein synthesis to the amount of protein in the feed has been reported in common carp by Jayaram and Shetty (1980 b). However, no such relationship was observed in the present study. The protein deposition values in the present study are in agreement with PER and protein digestibility co-efficients.

The fat content of the carcass basically determine the quality of fish. Because of its high energy value, it is also important in the evaluation of nutrient utilization of feed At the same time, among the nutrients, fat always shows the greatest fluctuation. Increased dietary lipid level enhances the deposition of fat (Buckley and Groves 1979, Reinitz and Hitzel, 1980). The same pattern of relationship has also been observed in the present study, the order of fat deposition being CM > SQM > PHM > FM > SYM which almost agree with the crude fat level in the corresponding feeds. However, Borthakur (1983) found no relationship between body fat deposition and fat content in feed. Dabrowski (1977) and Jauncey (1981) reported the fat content of the muscle increasing with a decrease in the protein content of the diet in common carp and grass carp respectively, while Luquet and Sabaut, (1973), and Ogino _et al_. (1976) noticed a reverse trend in gilthead bream and common carp respectively. But the above type of correlation could not be observed in the present study and fat content in the body was in agreement with lipid digestibility values.

Ash deposition shows a positive correlation with ash content present in the diets. The ash content observed in the present study is within the range reported by Hung et al (1980) in C. chanos.

An inverse relationship between protein and carbohydrate content of the feed was not found in the present study as reported by Khawaja (1966).

Variation in the calorific value of fish fed on different diets were between 93.23 and 121.7 Kcal/100 g fish. The energy content of the carcass varied mainly with the protein and fat contents of the muscle. Buckley and Groves (1979) also have reported a similar condition based on data obtained for various fish species. In the present study, the maximum value of energy content (121.7 Kcal/100 gm of fish) was found in fish fed on CM due to the high protein and lipid content in them Fairly good amount of increase in the energy content, was observed in fish fed on SQM. Calorific value of fish fed on PHM decreased by 2.67% indicating the poor quality of feed. This may be due to the lack of true protein in prawnhead meal. It can also be due to the increase in the moisture content of muscle by 0.87% and hence the decrease in NFE content by 1.7%.

5.3.3 Based on Oxygen consumption, Ammonia excretion and O:N ratio.

The increased oxygen consumption rate immediately after feeding is primarily due to the metabolism of amino acids. This increase is proportional to the percentage of protein in the diet. Ingestion of carbohydrates and lipid will elicit only a less oxygen consumption when compared to protein (Lovell, 1989). The oxygen consumption rate of fish fed on various diets shows the quality of protein to be in the order CM > SQM > FM > SYM > PHM.

Rate of ammonia production is dependent on the amount and quality (amino acid composition) of protein fed. If a protein of poor amino acid balance is fed, less protein synthesis per unit of ingested protein occur and the unused amino acids are deaminated and excreted as nitrogen (Lovell, 1989).

The comparatively lower rates of ammonia excretion by fish fed on CM, SQM and FM indicate that animal protein sources are superior in quality, for milkfish than plant source (SYM). The higher ammonia excretion rate of fish fed on PHM, although not significantly different from fish fed on SYM, indicate the poor quality of prawnhead meal as a protein souce. This may be because of its higher non-protein-nitrogen content. Ming (1985) and Kiron (1989) could observe an inverse relationship between ammonia excretion amd PER values. Such a relation was also observed in the present study.

The higher O:N ratios for CM (12.566) SQM (10.769) and FM (8.388) indicate that the protein from these sources are superior to *C. chanos* fry. The lower rate observed in fish fed on PHM (4.098) indicate increased amount of protein catabolism, which is also reflected in the low growth attained by the fish.

5.3.4 Based on Growth Studies.

The evaluation of supplementary feeds, based on survival rate, gain in the length and weight, and specific growth rate also reflect the same order of performance of feeds as observed in the efficiency of feeds. C. chanos fry fed with CM show superior performance followed by SQM and FM. Performance of fish fed on SYM was not satisfactory, while PHM was the poorest feed for C. chanos fry.

Hence it may be presumed that C. chanos fry utilizes feeds compounded from animal protein sources better than that from plant protein source. Samsi (1979) also reported animal protein sources like fish meal, meat and bone meal and shrimphead meal to be better utilized by C. chanos fingerlings than plant protein sources like soyabean meal, copra meal and ipil-ipil leaf meal. The studies by Kiron (1989) on Liza parsia fry also show a similar trend.

Reports on the use of clam meal and squid meal in finfish feeds are scarce. Borthakur (1983) has reported clam meal (20% of diet) along with silkworm pupae yield best growth in mrigal and common carp. Among the different protein sources used in the present study, the protein of clam meal may be of superior quality. Surya Narayana and Alexander (1972) have reported clam meat (Villorita sp.) as a good source of protein, having maximum number of free amino acids. The O N ratio of the present study also indicate that clam meal protein is best utilized for growth purposes than any other protein used in the study. Besides, the protein and lipid digestibilities of CM are the highest among the feeds. The highest stability of CM might also have contributed to the less leaching out of the nutrients in the feeds. These factors might have resulted in the better utilization of feed which resulted in the highest growth.

Squid meal contains high protein and ample amount of protein amino acids (Deshimaru and Shigueno (1972). A survey of literature based on the study of Deshimaru and Shigueno (1972), Lovell (1989) and Seneriches and Chiu (1988) shows that a combination of squid meal and groundnut oil cake (the ingredients used in the SOM feed) can provide, amino acids almost similar to that of milkfish fry. Squid meal also contains some natural feed attractors (Paulraj, 1989). Its higher digestibility of protein and lipid also have contributed to the better growth rate of C.chanos fry fed on SQM.

In the present study, performance of FM was only next to CM or SQM. Although Timbol (1969), Santiago et al. (1983), and Seneriches and Chiu (1988) have reported the superiority of fish meal in the feeds of C.chanos juveniles, none of these workers had tried yet another source, rich in protein like clam meal or squid meal instead of fish meal, in the diets they used. Kitamikado et al. (1964) have shown that rainbow trout of 10 or 100 g size digest white fish meal better than the fish of 6 g size Ferraris et al (1986) have also reported the digestibility of fish meal in C.chanos increasing from 2 g to 240 g size. C.cnanos fry, used in the present study was only 0.017 g in size. Probably subadults of milkfish can digest fish meal better than youngones. Further Steffens (1981) has mentioned that protein utilization depends on the size of fish, apart from species, protein quality, environmental factors etc. However, there are also reports of lower values of growth rate and protein utilization in rainbow trout raised on Peruvian fish meal (Nomura et al., 1973), and Lates calcarifer fry fed with fish meal from Thailand and Norway (Cho, 1985). Hence it seems that, quality of fish meal may also influence the growth performance in fish fed on them. Although fish meals from different places may have the same EAA composition, the biological availability of these EAA are not known and the chemical analysis of feed does not really indicate the quality of fish meal The Non-Protein Nitrogen (NPN) fraction and Total Amino Acids (TAA) may also vary among the fish meal (Cho, 1985) In the present study too, the biological availability of EAA, or the NPN or TAA content of the fish meal used are not known, as it was compounded from locally available Nemipterus sp The quality of the original raw fish, amount of small fish/fish head and tail used, processing technique, overheating during processing, method of storage, and level of ash content in the meal and oil also affect the quality of fish meal (Tan and Lee, 1975 Cho, 1985).

The growth performance of fish fed on SYM was 46.5% less than those fed on FM Similar results have also been obtained for common carp (Viola et al., 1982). Davis and Stickney (1978) reported a 32% reduction and Wu and Jan (1977) a 27% reduction in Oreochromis aureus, while Jackson et al. (1982) reported a 33% reduction in growth in O.mossambicus, when soyabean meal was used as a protein source instead of fish meal in diets, they used.

SYM feed used in the present study, contained only plant proteins Plant proteins are deficient in certain essential amino acids like lysine and methionine (N.R.C , 1983). Soyabean also contain trypsin inhibitors which contribute to poor utilization of protein (Smith, 1977). Although the soyabean meal used in the present study, was solvent extracted and autoclaved during cooking, it is not known, to what extent the antinutritional factors have been removed from it. The higher inclusion level (63%) of soyabean in the diet used in the present study, may also have contributed to the unsatisfactory performance of soyabean meal. According to Cho et al. (1985) higher levels (73%) of soyabean meal can influence the acceptability of feed by reducing feed intake significantly, despite its highly digestible nutrients. A low food intake (0.731 g /fish/42 days) of C chanos fry, when compared to fish fed on other feeds (0.783 to 1.138 g/fish/ 42 days) used in the present study, a high FCR and low protein and lipid digestibility all point out that higher level of inclusion of soyabean meal has affected the growth of C.chanos fry fed on this diet. It was also observed that the milkfish fry in the initial stages were found to feed poorly in SYM. Viola and Arieli (1983) have also observed a similar situation in tilapia when fed with soyabean meal. They attributed this to the inability of juvenile fishes to digest carbohydrate owing to their lesser developed digestive system. This may be true with C.chanos fry also Ferraris et al. (1986) have also reported that younger milkfish (2 g size) digest protein from soyabean meal less efficiently than adult fish. The poor utilization of plant proteins can also be attributed to the high crude fibre (mainly cellulose), which itself is hardly digested, and may also envelop and protect other, more digestible nutrients such as protein and carbohydrates from digestive enzymes (Hepher, 1988).

PHM is not a good feed for C chanos fry as revealed by the poor growth, FCR and biochemical composition of fish. This may be because of the low content of true protein in prawnhead waste. Anil (1981), Srikanth (1986), and Kiron (1989) have also observed the poor performance of shrimphead waste in fishes Prawnhead waste contains high amount of chitin (contributing to about 10 to 15% of total nitrogen- ie., NPN)(Lovell, 1989), which is intimately associated with Ca salts and pigments. (Lindsay et al., 1984) Prawnhead waste contains 78 7% digestible proteins, rich in methionine and lysine. But its utilization may be hampered by the Ca and chitin present in it (Meyers and Rutledge, 1971, Simpson et al., 1981). Chitin is a non-soluble particulate fibre, contributing to the greater particular bulk of feed (Nomani et al , 1979), and majority of the monogastric animals including fish do not have the intestinal flora to digest it (Stickney and Shumway, 1974) Robinette and Dearing (1978) attributed the lower performance of shrimphead meal to the lower digestibility or palatability or to the deficiency of some unidentified growth factors. In the present study too, the digestibility of protein was lowest for PHM, although lipid digestibility was fairly good These factors may also have hampered the growth of C.chanos fry fed on PHM diet.

However, there are reports of better growth performance when shrimphead meal is incorporated at lower levels in fish feeds Supplementing fish meal with shrimphead meal at 8% level of dietary protein has promoted good growth and survival of milkfish fry (Alava and Lim, 1988). Shrimphead meal promotes fish growth when present in feed at 5% level and inhibits growth if incorporated at 20% level in rainbow trout (Fowler and Banks, 1976). According to Kiron (1989) the quality of shrimp waste meal determines, how best it can be included in the diets of Liza parsia fry. So it seems, that both quality and quantity of shrimphead meal are determinant factors regarding its incorporation in the diets of fishes.

5.4 Optimum Food Ration

In the present study, the survival rate of C.chanos fry (0.107 g/2.0 cms) fed at 3% or 12% of body weight was not significantly different. However, specific growth rates were significantly lower at lower levels of feeding. Chiu et al. (1987) have reported significant increase in growth when feeding level was increased from 5 to 9% in C.chanos juveniles. In the present study, maximum gain in weight was obtained when fish were fed at 12% of body weight. But, feeding beyond 15% showed even a slight reduction in the weight gain. Food conversion rate was however, best at lower levels of feeding. A similar trend was also observed by Kiron and Paulraj (1988) in Liza parsia fry. Graphical extrapolation shows that feeding the fry at 5.2% of body weight to be optimum for C.chanos fry. The maintenance ration is at 1% of the body weight and the maximum ration at 12% of the body weight.

Earlier workers have indicated feeding fish beyond a certain level is wasteful in terms of feed cost and water quality maintenance. Ghosh et al. (1984), and Kiron and Paulraj (1988) have reported that it is better to feed common carp and *Liza parsia* fry, respectively at 4% of body weight. In *Epinephelus salmoides* the maintenance, optimum and maximum rations are reported to be at 1.4%, 5% and 9% of body weight respectively (Chua and Teng, 1982). In the present study, when survival and FCR are also considered, it can be concluded that the optimum food ration of *C.chanos* fry, fed with clam meal diet containing 40% crude protein, is 5.2% of the body weight, at a feeding frequency of two times a day (9 a.m. and 4 p.m.) and at a salinity of 4 to 6 ppt.

SUMMARY

6 SUMMARY

The present study was conducted to evaluate the efficacy and keeping quality of supplementary feeds compounded from five different protein sources viz., fish meal, squid meal, clam meal, prawnhead waste meal, and soyabean meal, for the nursery rearing of Chanos chanos fry. The optimum food ration of the selected feed which gave the best performance of feed efficiency in growth and survival among the five feeds was also studied.

1. The proximate analysis of ingredients used in the formulation of feeds revealed, that squid meal and fish meal had higher crude protein content (80.48 and 70 22% respectively) than clam meal (50.62%), prawnhead waste (50.15%) or soyabean meal (47.6%). Clam meal had the highest crude fat content (11.1%), while the crude fat content of prawnhead waste meal, squid meal and fish meal were 6.9, 6.5 and 5.9% respectively.

2. The crude protein content of all the five formulated feeds were around 40%, the range being 39.78 (CM) to 40.44% (SQM). The crude fat content was the highest for CM (8.1%) and the lowest for SYM (5.8%). The crude fibre content of feeds ranged from 3.59% (CM) to 8.8% (SYM). The NFE content was the highest in FM (26.7%) and the lowest in PHM (20.6%). The ash content was the highest for PHM (17.04%). The feeds were almost isocalorific and their energy content ranged from 3 13 to 3.33 Kcal/g.

3. The moisture and NFE contents of the formulated feeds showed an increase, while crude protein, fat, crude fibre and ash showed a decrease in their contents after four months of storage. However, there was not much variation in their contents showing that the quality of foods formulated was satisfactory. The sinking rate of the food also increased after four months of storage.

4. The most stable feed both before and after storage was CM, followed by SYM, SQM, FM and PHM.

5. The variations observed in the water quality parameters were found to be well within the tolerance limits of milkfish fry indicating that supplementary feeds had no adverse effect on the quality of water.

6. The FCR of fish fed on various supplementary feeds for 42 days showed that fish fed on animal proteins, as chief protein source had better food conversion ratios than those fed on plant protein source. The FCR of CM, SQM, FM, SYM and PHM were 1.74, 2.94, 3.65, 5.32 and 10.9 respectively.

7. The PER and protein digestibility co-efficient values have shown that protein from clam meal is better utilized for growth purposes than the protein from squid meal, fish meal, soyabean meal or prawnhead waste meal. The PER and protein digestibility values for fish fed on CM were 1.45 and 97.34% respectively.

8. Lipid digestibility values indicate that lipid from animal protein sources is better utilized than that from plant source. Thus fish fed on CM showed the highest lipid digestibility (98.08%), while fish fed on SYM

showed the least (47.49%).

9. The biochemical composition of fish showed maximum synthesis of protein and deposition of fat in fish fed on CM, followed by SQM, while the least protein synthesis was in fish fed on PHM. Ash deposition has shown a positive correlation with the ash content of the diet.

10 O N ratio was found to be significantly influenced by the chief protein source used in the feed The O N ratio was maximum (10.769), oxygen consumption rate fairly high (0.5405 mg O_2/g/hr) and the ammonia excretion rate minimum (0.043 mg NH_3 N/g/hr) in fish fed on CM.

11. The evaluation of supplementary feeds based on survival rate, gain in length and weight and specific growth rate attained by fish after 42 days of rearing indicate the superior performance of animal protein sources (except prawnhead waste meal) compared to plant source. The highest gain in growth was obtained for fish fed on CM followed by fish fed on SQM and FM. Performance of fish fed on SYM was not satisfatory, while PHM was found to be a poor feed for C chanos fry.

12. The optimum food ration of C. chanos fry fed with clam meal diet, containing 40% crude protein is 5.2% of the body weight, at a feeding frequency of two times (9 a.m. and 4 p.m.) a day and at a salinity of 4 - 6 ppt.

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EVALUATION OF SUPPLEMENTARY FEEDS AND OPTIMUM RATION FOR *CHANOS CHANOS* (FORSKAL) FRY

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ABSTRACT

The present study was conducted in *C. chanos* fry, to evaluate the efficacy and keeping quality of supplementary feeds compounded from five different protein sources viz., fish meal (FM), squid meal (SQM), clam meal (CM), prawnhead waste meal (PHM) and soyabean meal (SYM). All the feeds used in the study were isocalorific (3.13 - 3.33 kcal/g) and isonitrogenous (39.78 - 40.44% crude protein).

The quality of feed after four months of storage was satisfactory as there was not much variation in their nutrient contents.

The FCR of fish fed on various supplementary feeds for 42 days were 1.74, 2.94, 3.65, 5.32 and 10.9 for CM, SQM, FM, SYM and PHM, respectively. The PER, protein digestibility co-efficient and O:N ratio indicate that protein from clam meal is better utilized for growth purposes than those from others. The PER value ranged from 0.23 (PHM) to 1.45 (CM). The protein digestibility co-efficient for CM, SQM, FM, SYM and PHM were 97.24, 91.86, 86.56, 77.78 and 65.23%, respectively. The O:N ratio was highest (10.769:1) for fish fed on CM. It seems that *C. chanos* fry digest lipid from animal protein sources better (digestibility values ranging from 84.17% (PHM) to 98.8% (CM)) than that from plant source SYM (47.49%). The biochemical composition of fish showed maximum synthesis of protein and deposition of fat in fish fed on CM, followed by SQM, and the least in fish fed on PHM.

The evaluation of supplementary foods based on survival rate, gain in length and weight, and specific growth rate indicate the superior performance of animal protein sources (except prawnhead waste meal) to plant source. The highest gain in growth was attained by fish fed on CM, followed by SQM and FM. Performance of fish fed on SYM was not satisfactory, while PHM was a poor feed for C. chanos fry.

The optimum food ration of fry is at 5.2% of the body weight when fed with CM, twice a day, at 4-6 ppt salinity.