DISCUSSION
4. DISCUSSION

The quantitative requirement of any food depends largely on its composition. The most efficient level of feeding in aquaculture is attained only when the correct supply of energy and essential nutrients are available in the proportions required by the fish for maintenance and growth (Hepher, 1988). Any deviation from this ideal composition will change the quantitative food requirement and ultimately affect the growth of the fish. It is also well known that food consumption of aquatic thermoconformers is significantly influenced by various extrinsic (eg. Temperature and salinity: Brett and Higgs, 1970., Brett, 1971., Tyler and Dunn, 1976., Bryant and Matty, 1981., De silva and Gunasekara, 1989., Condrey, 1982; Food quality : Carefoot, 1967., Vivekanandan and Pandian, 1977., Mathavan et al., 1976) and intrinsic factors (eg : Body size : Pandian, 1967., Garber, 1983; Hunger level : Elliot and Persson, 1978., Jobling and Davies, 1979., Jobling, 1980, 1986).

The present results showed that for *M. cephalus* the optimum protein level varied from 35 to 40%. Maximum percent growth increase, specific growth rate, food conversion efficiency and food conversion ratio were recorded at 40% protein level. The productive protein value of *M. cephalus* was relatively high at the highest protein level (40%) and very low for those individuals fed with other protein level (25, 30 and 35%). It is evident therefore that the optimum dietary protein content for *M. cephalus* would be of 40% when compared with (40 to 45%) that recorded for cichlids (Jauncey and Ross, 1982). Delong et al. (1958) have also reported the same level (35 to 40%) of optimum protein for better growth and food conversion in Chinook salmon trout and further increase in dietary protein to 50%, the specific growth rate and food conversion efficiency
decreased. Jauncey and Ross (1982) also observed that *Sarotherodon mossambicus* did not grow any better when dietary protein level had increased above a certain level. Similar observations in *Chanos chanos* were also reported by Lim *et al.* (1979) to that of *M. cephalus*. Gopal *et al.* (1999) reported that for the mullet species *Liza macrolepis* which was fed with casein – gelatin based isocalorific purified diets with graded levels of protein (10-60%) showed that the optimum dietary protein required for better relative growth rate, specific growth rate and survival was 40%. Similar to the early observations Venkadesh and Palavesam (1999) also pointed out that the optimum protein requirement for mullet species is 40% and this is in inconsistence with the findings of the present observations. In another brackish water fish *Etroplus suratensis* Jayaprakash and Sunil Kumar (1999) observed better growth at 35% protein level and thus recommended that a diet with 35% protein is optimum for maximum growth and high feed utilization. Das and Ray (1991) reported that in the *Cirrhinus mrigala* also, the optimum dietary protein level was 35% and the food conservation ratio increased with increasing dietary protein up to 35% and thereafter decreased, being maximum for the diet with 55% protein. Gunasekaran *et al.* (1995) reported the best growth for *O. niloticus* (3g) fed at 40% protein diet compared with those fish fed 10, 17 and 25% protein diets.

Though protein is the major constituent in the fish feed, its requirement is mainly determined by the availability of non-protein energy source in the diet. In the present study at the tested protein level (25 to 40%) with increase in carbohydrate or lipid level, the growth performance i.e. percent growth increase specific growth rate and feed conversion efficiency of *M. cephalus* also increased. This increase was much more effective for those fish reared on low protein diets (25 and 30%) than that of those reared on high protein diets (35 and 40%). At
constant protein level, with increase in non-protein nutrients the fish derive energy for maintenance and growth from the latter nutrients as the result of the sparing action. This may be the reason for the better growth performance displayed by *M. cephalus* fed on non-protein nutrient supplemented diets in the present study. Additionally the less variation or attaining a level of saturation in growth performance at high protein diets (35 and 40%) fed fish still implies that the deriving of more metabolizable energy from the protein part of the diet and the resultant catabolic energy loss may be attributed to the low disparity in growth performance at high protein diet (40%) supplemented with either carbohydrate or lipid. The growth performance data presented here for *M. cephalus* fed on carbohydrate and lipid supplemented diets agree well with Ahamad Ali (1982), who reported the enhanced growth in prawn with respect to increase in dietary carbohydrate. He also pointed out that this may be due to the protein sparing action of carbohydrate in which the increased level of carbohydrate in the diet might have provided larger quantities of energy required for the metabolic process of the animals while more and more protein had been spared for growth. The enhanced growth of *M. cephalus* which received higher level of carbohydrate or lipid supplemented low protein diets may be attributed to obtain metabolic energy both from protein and non protein nutrients accounting for growth elevation.

The present result also showed that, irrespective of the dietary protein level, food consumption and conversion efficiency of *M. cephalus* was increased with increase in carbohydrate level and decreased with increase in the protein content in the diet. The increase in feed conversion efficiency was 10.93, 14.96, 10.60 and 16.53% over those fish fed with diet devoid of carbohydrate at 25, 30, 35 and 40% protein level reared at 15ppt salinity. In a series of investigations, Jobling (1980, 1981) demonstrated that the return of appetite and feeding rate of the fish
*Pleuronetes platessa* depends on the quality of food as well as gut evacuation time. The duration for which the food is retained in the gut determined feeding rate. Shorter the duration of food retention in the gut, higher is the feeding rate. It was also established that when high content of carbohydrate was added in the diet, the undigestable part of the carbohydrate move at a faster rate in the alimentary canal resulting in the higher feeding (Hepher, 1988). The maximum feeding rate and feed conversion efficiency recorded for *M. cephalus* fed with high carbohydrate diet may be attributed to the above said fact. Similarly the food consumption of *M. cephalus* showed an increase of 11.03, 19.66, 18.60 and 15.30% over those fish fed with diets devoid of the lipid at 25, 30, 35 and 40% protein level reared at 15ppt salinity. This present trend also confirmed the earlier observation of Jobling (1980 and 1981) and Hepher (1988), that the enhanced feeding rate of fish accomplished by the availability of greater portion of undigestible non-protein energy sources in the diet. This is mainly due to the fact that the fish try to compensate the poor quality diet by enhancing the feeding rate and this is true for *M. cephalus* fed with both carbohydrate and lipid diets in the present study. Ruohomen *et al.* (1999) reported that in rainbow trout the tendency of increased dry matter intake with increasing consumption of carbohydrate might be the result of increased gastric emptying and faster return of appetite in this fish; since starch is known to accelerate the passage of food through intestine of rainbow trout (Spannhof and Platikow, 1983).

The variation in food consumption of *M. cephalus* fed with varying protein diets in the present study is analogous with the finding of Tabachek (1986) for Artic charr; Wang *et al.* (1985) for *Tilapia nilotica* and Beena somnath (1991) for *Labeo rohita* and *Catla catla*. Although digestive and metabolic systems of fish are known to be better adapted to use of protein and lipid than carbohydrate for
energy, some warm water species, particularly herbivorous and omnivorous digest and metabolize carbohydrate relatively well. NRC/NAS (1983); New (1986) and Cowey (1988) have reviewed the use of dietary carbohydrate and its influence on fish growth. The optimum levels of dietary carbohydrate for most fishes are reported to be within the range of 10 to 30% (NAS – NRC, 1983). Furuichi et al. (1988) reported that carps grow well even on diets containing low levels of dietary carbohydrate. In general warm water fishes do not have true carbohydrate requirement (NAS/NRC – 1981). But incorporation of a certain level of carbohydrate in the diet influences conversion efficiencies and overall growth of fish. The effect of dietary carbohydrate level in fishes were studied by Brauge et al. (1994) and Kim and Kaushik (1992) in rainbow trout, Erfanullah and Jafri (1993) in Labeo rohita; Nagabhushanam et al. (1988) in Macrobrachium kustinesis, Ahamad Ali (1996 and 1993) in Peneaus indicus, respectively.

The percentage increase in specific growth rate of M. cephalus fed on carbohydrate supplemented diets at 25 to 40% protein levels was 102.8, 140.88, 138.68 and 126.10%, when compared with those fish fed with diet devoid of carbohydrate. Moreover, the increase in growth responses over the control was much more pronounced at low protein diet (30 and 35%) fed fish than those fish received high protein diet (40%). At higher protein level (40%), high carbohydrate content did not show any remarkable influence on the growth performances of M. cephalus. The present result finds support from the findings of Hemre et al. (1995a, 1995b) and Aksnes (1995) for Atlantic salmon. They pointed out that the growth and food utilization of Atlantic salmon was impaired when the starch content was increased from 220 to 310 g Kg\(^{-1}\). But no significant effects on growth have been detected at lower inclusion level ranging from 50 to 230 g Kg\(^{-1}\). In contradiction to this, Kaushik et al (1989) stated that the inclusion of high level
of carbohydrate does not adversely affect the growth performance or retention of major nutrients in rainbow trout. The role of carbohydrate in fish diet has not received as much attention from researchers as has protein and lipid because, unlike protein and lipid, carbohydrate is not considered as an essential nutrient in diet. Its deficiency does not cause any disease. Nevertheless carbohydrate is an important and less expensive source of energy than any other energy component in the diet.

In the present study *M. cephalus* fed on lipid supplemented diets also showed an increasing trend of growth responses at the tested protein level. The increase in percent growth increase and specific growth rate of *M. cephalus* was 363.38, 189.65, 189.19 and 211.76% and 308.07, 148.25, 161.30 and 161.69% respectively in 25, 30, 35 and 40% protein levels over those fish fed with control diet at 15 ppt salinity. The increase in growth responses over the control diet fed fish was more obvious at low protein level (25%), similar to that recorded for those fish fed on carbohydrate supplemented diets.

Many researchers have studied the dietary lipid requirements for optimal growth in warm water fishes. Johnsen and Wandsvik (1991); Kiaerskou (1991) and Heinen et al. (1995) reported that the salmonid diets, which are easily digestible largely because of higher fat content, could be used to achieve better food conversion and reduce production of wastes. Heinen et al. (1995) also reported that in *Oncorhynchus mykiss*, a diet with 18% fat and 45% crude protein diet showed significant weight gain than that fed with 12% fat and 38% protein diet and with 17% fat and 45% protein diet. Likewise in walking catfish *Clarias batrachus*, Anwar and Jafri (1995) reported that the dietary lipid concentration of 7 to 9% was optimum for those fish fed with 40% protein at iso-caloric energy
levels. They also reported that increase in dietary lipid also resulted in greater accumulation of liver lipid; whereas, the hepatosomatic index did not vary much. Tibaldi et al. (1996) has done considerable works on the energy requirements in Dentex dentex particularly on the protein to lipid ratio at varying protein levels. They concluded that the growth performance was lowest in fish given diet containing 44.4 or 49.5% protein and 12% lipid and was improved either with diet supplying 55.8 or 49.3% protein and 17.4% lipid. But, with regard to protein efficiency, they recommended 44.3% protein and 17.2% lipid was suitable for the juvenile dentex.

While doing experiment on hybrid striped bass Nematipour et al. (1992) reported that the fish were able to utilize carbohydrate efficiently for energy and that lipid deposition through dietary lipid in various tissues could be partially replaced with carbohydrate to improve fish quality and productivity. Takeuchi et al. (1978(a, b, c) and Watanabe, 1977) evaluated protein to lipid ratio of 35%: 18% in diets for juvenile rainbow trout as optimal, provided high quality protein and fat are used. Similarly, in the present study M. cephalus displayed maximum growth performance at 40% protein level with 15% lipid level. This is in accordance with the results reported by Heinen et al. (1995) for rainbow trout (O. mykiss). From this present findings it could be observed that M. cephalus required still higher protein: lipid ratio when compared with rainbow trout.

The factors, which alter the growth performances in fishes, will also exert their influence on body composition. Accordingly, the body composition of fish is influenced by several factors including individual variation, strain, sex, age, size, dietary protein and digestible energy concentrations, dietary energy and protein ratio and feed allowance. The present results showed that next to growth
responses, body composition of *M. cephalus* was significantly influenced by dietary treatment. The carcass protein and carbohydrate contents were increased with increase in dietary carbohydrate level, whereas, the lipid content decreased with the increase in the carbohydrate level in the diet. Likewise in lipid supplemented diet fed fish, the protein and lipid contents increased with respect to increase in lipid level in the diet. On the other hand the carbohydrate deposition decreased with increase in lipid level in the diet. These differences were also obvious in faecal biochemical composition. The results of the present study are in general agreement with several previous reports that reveals that the feeding of high fat diets tend to result in fattier fish (Hoard, 1992; Hillestad and Johnsen, 1994; Koskela, 1995; Wathne, 1995; Koskela et al., 1998). Thus deposition of body fat may be influenced by dietary composition.

Degani *et al.* (1986) were the first to examine the effect of dietary carbohydrate source on growth and fatty acid composition. This effect may help to understand how the fish can use different carbohydrates as a source of energy. The pattern of changes in body carbohydrate content of *M. cephalus* fed on carbohydrate supplemented diets in the present study showed that the maximum carbohydrate accumulation was registered for those fish fed with 40% protein diet with 20% carbohydrate. It implies that a maximum of 20% carbohydrate in the diet is essential for the efficient utilization of 40% protein. While studying the effect of carbohydrate source on fingerlings of *Labeo rohita* Erfanullah and Jafri (1995) reported a higher amount of carcass carbohydrate content when fed on diet containing 30% sucrose and 40% crude protein. Changes in whole-body composition in Nile tilapia *O. niloticus* with respect to protein level in the diet was also reported by Al Hafedh (1999). *i.e.* lower percentage of protein and higher
lipid level in fish fed with low protein (25 to 30%) than those fish fed with 40 and 45% protein diets.

Dietary lipid levels, as well as fatty acid composition have been shown to affect the chemical composition of the farmed Atlantic salmon muscle, which is important with respect to product characteristic such as taste and nutritional quality (Hardy et al., 1987; Thomasen and Rosjo, 1989; Waagbo et al., 1993). Feeding very high lipid diet may impart a strain on fish with respect to the oxidation in biomembranes. The influence of dietary lipid quality on body lipid composition was also demonstrated by Cowey et al. (1975); Garling and Wilson (1976), Kalogeropoulos et al. (1991), Argyropoulos et al. (1992), Krajnovic – Ozretic et al. (1994); Garcia Gallego and Akharbach (1998) for various fish species. Low protein and high lipid deposition in low protein diets (25-30%) fed O. niloticus was also reported by Al-Hafedh (1999) than those fish fed with high protein diets (40 and 45%). White sturgeon fed with maltose or glucose diets (Hung et al., 1989) and Tilapia (Tung and Shiau, 1991) fed with glucose, dextrin or starch diets had a higher rate of body lipid accumulation.

This lipogenesis may be ascribed to higher lipogenic activities in fish. The influence of dietary lipid on carcass content was also reported by Hossain et al., (1998). An increase in fattiness with increase in dietary energy : protein ratio was also reported in channel catfish (Li and Robinson, 1999) Increased lipid deposition in muscle was also noticed in fishes like rainbow trout (Lee and Putnam, 1973) channel catfish (Garling and Wilson, 1977; Santha and Galtin, 1999), red drum (Williams and Robinson, 1988; Ellis and Reish, 1991; Serrano et al., 1992) and Nile tilapia (Hanley, 1991), when fed with diets containing higher level of lipid.
Studying carbohydrate: lipid ratio on hybrid striped bass Nematipour et al. (1992) reported a marked increase in whole body dry matter and lipid contents and a decrease in protein content. Similar trend was also reported on striped bass (Millikin, 1982) fed with high lipid diets. Variations in whole body composition of *M. cephalus* observed in the present study could be explained by the significant differences in the amount of lipid accumulated in the body. Here a steady increase in body protein of *M. cephalus* was caused by addition of both carbohydrate and lipid in the diets. The utilized carbohydrate and lipid at the tested dietary protein levels is deposited in the muscle as protein source. The increased protein content in the muscle of *M. cephalus* fed diet containing 40% protein with 20% carbohydrate indicated a better protein synthesis and growth. From this, it is evident that a diet with 40% protein having 20% carbohydrate is optimum for maximum protein deposition as seen in the case of lipid or carbohydrate. The present observation is in tune with the findings of Hossain et al. (1998). They reported that the protein: energy ratio in the diet significantly influenced the carcass composition of *Puntius gonionotus* than the dietary protein level. Similar finding was also reported by Murai et al. (1985) for carp fingerlings. Replacement of feed protein by carbohydrate or fat initially increases the protein conversion efficiency, but ultimately consumption will decrease as the fish become fatter and this consequently will lead to a decrease in the maximum gain. This effect seems to be more pronounced for an increase of carbohydrate than for an increase of fat content in the diet.

In the present study the body composition of *M. cephalus* fed with high carbohydrate and low protein diet did not show any reduction in body protein. High carbohydrate level thus helped in sparing dietary protein at low protein levels. Such protein sparing action of dietary carbohydrate was also reported in
Penaeus setiferus (Andrews et al., 1972), P. duorarum (Sick and Andrews, 1973) and P. indicus (Ahamad Ali, 1982). The beneficial effect of protein sparing nutrient has also been studied (Viola and Rappaport, 1979; Cho and Kaushik, 1985). In contradiction to this the whole body protein concentration of red drum (Sciaenops ocellatus) did not show significant difference as dietary protein or carbohydrate and this is mainly due to the non formation or non-occurrence of protein sparing effect of carbohydrate or lipid among the dietary treatments. These results clearly indicate that the protein sparing effect of non-protein energy sources may vary from species to species and this sparing effect was much more established by altering the carcass composition among experimental diets, in the present study.

Moreover, in fishes the growth performance and biochemical constituents i.e. nutrient deposition is mainly governed by digestibility of the feed stuffs being consumed by the fish. The digestibility of the feed stuffs in turn is influenced by physical and biological parameters such as temperature, water quality, fish size, age and feeding frequency as demonstrated in various fish species, both marine and fresh water such as Oreochromis mossambicus and Scophthalmus maximus (Windell and Brown 1978; Jollivet et al., 1988). The source and composition of feed stuffs also bring about significant variations in digestibility as described in O. aureus and O. niloticus (Degani and Yehuda, 1999). The determination of digestibility together with chemical analysis allows more exact estimation of the natural value of a given nutrient source (Plakas and Katamaya, 1981).

The present results on nutrient consumption, digestibility and nutrient retention of M. cephalus showed great variation corresponding to variation in protein - carbohydrate and protein - lipid ratios in the diets. Here, with increase in
dietary carbohydrate the protein and carbohydrate digestibility decreased. But for *M. cephalus* fed with diets containing 20 to 40 % protein diets with 0 to 15% lipid, the protein and lipid digestibility increased; whereas, carbohydrate digestibility decreased. The result of the present study indicates that the digestibility of the given component is affected by the remaining components of the diets. The range of protein digestibility obtained in the present study is in agreement with the reported values of 90.2% (Kim, 1974) and 95% (NRC, 1977) for carp. The average protein digestibility ranged between 73.9 and 94.4% for mrigal fingerlings and between 71.9 and 75.4% for grass carp fingerlings. Brown *et al.*, (1985) also observed high protein digestibility (92%) for corn gluten in yearling of channel catfish.

Georgopoulos and Conides (1999) showed that in *Sparus aurata* there was a slight trend for decreased protein digestibility with increasing levels of starch in the diets. Same profile has also been observed in *Oncorhynchus mykiss* (Kitamikado *et al.*, 1964a; Inaba *et al.*, 1963). Rychly and Spannhof (1979) stated that any decrease in protein digestibility is attributed to the lower protein levels in the experimental diets in order to allow increased levels of starch in feeds and avoid any sparing effect.

The enhanced protein digestion of *M. cephalus* fed with carbohydrate added diets in the present study may be due to the acceleration in the proteolytic enzymes in the fish. According to Falge *et al.* (1978) although the increase in carbohydrate content of the diets actually reduces the activity of the proteolytic enzymes, this activity remains still quite high. It is doubtful therefore, whether the digestibility of the protein is really affected by carbohydrate in this way. In contradiction Tunison *et al.* (1943, 1944) have shown that the digestibility of protein by trout
depends on the content of carbohydrate in the feed, the higher the carbohydrate content, lower the protein digestibility. They explain that the undigested portion of the carbohydrate passes more rapidly through alimentary canal, carrying with it some of the proteins.

In the present study the experimental diets were formulated with varying protein level from 25 to 40% and the low protein levels may be the reasons for the high protein digestion and utilization by *M. cephalus* as the carbohydrate content of the diet increased. This was also well reflected in obtaining the higher protein production by *M. cephalus* fed on test diets. The beneficial effect of the incorporation of protein sparing nutrients will mainly depend on the ratios between the protein and energy as suggested for many fish species (Cho and Kaushik, 1985 and Viola and Rappaport, 1979). It was also found out that relatively cheap energy – yielding nutrients such as fat or carbohydrate have been found to reduce the oxidation of protein, satisfying the energy requirement of fish and thereby improving the utilization of the diets.

Digestibility of starch varies with fish species (Shimeno *et al.*, 1977), feeding level, level of inclusion in the diet (Inaba *et al.*, 1963; Singh and Nose, 1967; Chiou and Ogino, 1975; Bergot and Breque, 1983) and heat treatment of the raw material (Chiou and Ogino 1975; Jollivet *et al.*, 1988; Takeuchi *et al.*, 1990). Preliminary data on corn starch digestibility by gilthead bream suggest that it could be relatively high (Georgopoulos, 1990). However, starch digestibility varies greatly with its botanical origin for rainbow trout (Bergot, 1993) and this might be one of the factors affecting the digestibility of starch and this invariably improves with heat treatment (Bergot, 1993). It could be suggested that further
heat treatment of plant materials exhibiting good protein digestibility values might improve their nutritional value for gilthead bream.

Fibre is considered to be completely indigestible by fish (Kirchgessner et al., 1986 and Bergot, 1981). Furthermore, the existence of high levels of fibre could also interfere with the digestibility of other nutrients (Anderson, 1985; Buddington Hilton, 1987), thereby reducing the overall digestibility of the diet. The carbohydrate digestibility of the M. cephalus showed an increasing trend with increase in carbohydrate level in the diet, and it was well established at low protein diet (25-30%) fed fish than those fed with high protein diet (35 to 40%). Ufodike and Matty (1983) have also pointed out the high protein digestibility with the inclusion of digestible carbohydrate in the diet for carp Cyprinus carpio. The less differences in carbohydrate digestibility at high protein diet may have been in part due to high protein content (See also Shimeno et al., 1978).

Further, nutrient absorption depends on the time at which nutrients are in contact with the absorptive epithelium. The influence of dietary fibre on the movement of nutrients along the gastrointestinal tract is likely to influence nutrient absorption significantly. Dietary fibre can be divided into two categories: namely, viscous or water-soluble polysaccharides and non-viscous or water-insoluble polysaccharides. Water-soluble dietary fibres such as guar gum and pectin have been reported to delay stomach emptying in a number of studies (Holt et al., 1979; Wilmhirst and Crawley, 1980; Ebihara et al., 1981; Leeds et al., 1981; Schwartz et al., 1982; Tadesse, 1982). The delay has been attributed to an increased viscosity of the test meal (Wilmhirst and Crawley, 1980; Ehrlein and Prove, 1982) which might influence the absorption rate of
carbohydrate by fish. The fibre source used for the present experiment was cellulose. Cellulose is a water insoluble dietary fibre, which has been reported to increase stomach-emptying time in rainbow trout (Hilton et al., 1983). The increased gut emptying time by the cellulose added in the experimental diets may be the reason for the enhanced protein and carbohydrate digestion.

There have been a number of attempts to spare the use of protein as the main source in diets for farmed fish. Most of the researches involved in salmonids are focussed on increasing lipid levels in order to reduce the protein content of the diets at a constant energy level (Watanabe et al., 1979). They also reported that an increase in lipid content of feed from 5 to 23% increased the digestion of total energy by rainbow trout. This was due to an increase in the digestion of protein from 98.4 to 98.9% of carbohydrate from 50.7 to 58.5% and also that of the lipid themselves from 74.7 to 87.5% (Watanabe et al., 1979). This said fact also holds good for the present study. Here, *M. cephalus* fed with diets having 25 to 40% protein with 0 to 15 lipid showed an enhanced protein and lipid digestion themselves. Fish can usually absorb and utilize large amounts of fats in their diet (Yu et al., 1977; Reinitz and Hitzel, 1980). Lovern (1951) pointed out that brook trout can thrive on a diet containing 57% fat. Other authors have also reported that very high levels of dietary lipid were fed to fish without any growth depression or pathological effects. Thus rainbow trout were fed without any harm in diet containing 25 – 30% fat (Higashi et al., 1964. Kitamikado et al., 1964b).

In the present study the ability *M. cephalus* to utilize high level of lipid at low protein diets (25 to 30%) in turn spare dietary protein for growth. In many fish species, protein retention was improved by partly replacing dietary protein by lipids. Such protein-sparing effect have been demonstrated in salmon
(Garcia et al., 1981; Johnsen et al., 1991) trout (Beamish and Medland, 1996), carp (Watanabe et al., 1987) hybrid striped bass (Nematipour et al., 1992), yellow tail (Shimeno et al., 1980) red sea bream (Takeuchi et al., 1991) and in gilthead sea bream Sparus aurata (Vergara et al., 1996). Their results clearly suggest that nitrogen excretion resulting from protein catabolism for energy content of the diet decreases with decreasing the digestible protein to digestible energy ratio. However, despite the environmental interest, such high energy diets generally lead to greater fat deposition (Watanabe, 1982) which can reduce the commercial value of the product.

According to Marais and Kissil (1979) 9% of fat in a diet containing 44% protein would represent the maximum amount needed for optimum growth of S. aurata. Increase in efficiency of energy retention with increase in dietary lipid from 15 to 21% was also reported in gilthead seabream, Sparus aurata (Santianá et al., 1999). They have also reported a higher protein retention efficiency with diets containing 47 and 51% crude protein with high lipid content from an experiment on gilthead seabream fed diets with graded levels of crude protein (42 to 58%). Vergara et al. (1996) concluded that dietary protein level should be decreased from 58 to 40% when increasing the lipid content of dry matter from 9 to 15%. Despite this protein sparing effect, these authors did not find any significant effect of dietary lipid level on the Protein Efficiency Ratio (PER). Conversely, they observed a decrease in PER with the increase in protein in the diet. In the present study the maximum PER and Productive Protein Value (PPV) were obtained for M. cephalus fed with 40% protein with 15% lipid diets. Significant influence of both dietary protein and lipid (energy) on PER and PPV demonstrated the importance of these variables on protein utilization in fish. Trend in PER recorded in the present study clearly indicates that both protein to
carbohydrate and protein to lipid ratio of the diet influence the protein production in *M. cephalus*. This points out the beneficial effect (sparing) of sufficient carbohydrate or lipid incorporation in the diet of *M. cephalus*. This was consistent with the results obtained for other fish species (Bromley and Adkins, 1984). A linear increase in PER with increasing dietary energy ($r = 0.89; P < 0.05$) and decreasing cellulose in the dietary content of *O. mossambicus* was also reported by El Dahhar and Lovell (1995). When the carbohydrate or lipid content of the diet increases, it spares the protein store in the fish tissues. Parazo (1990) and Shiau and Huang (1990) also reported the positive increase in growth rate of Mozambique tilapia with increase in P: DE ratio in isocaloric diets.

Although the pattern of changes in PER in relation to dietary energy (from carbohydrate and lipid) shows similarity with the observations of De Silva *et al.* (1992) on Nile tilapia, it contradicts the findings of Reinitz *et al.* (1978), Watanabe *et al.* (1979), Tabachek (1986) and Shiau and Huang (1990), who noted a positive linear relationship between dietary energy (from lipid) and protein utilization. In general, the PER decreased with increasing weight of fish and dietary protein level. Jauncey (1982) and De Silva *et al.* (1989) also reported FCR and PER decreasing with increasing dietary protein content. A decrease in PER with increase in dietary protein level was also reported by Vergara *et al.* (1996).

In contradiction to this, the dietary protein level showed a positive influence on the PER of *M. cephalus*. In the present study the diets were formulated at the same protein to energy ratio. This isoprotein to energy ratio in higher protein diets may support to utilize more protein and thereby increase the PER of *M. cephalus*. Dabrowski (1979) also reported different patterns of changes in PER in relation to dietary protein level and found out that the relationship
between dietary protein and PER differs from species to species. From this it may be inferred that to utilize the dietary protein more efficiently, sufficient energy in the diets is of more importance. Adequate energy in the form of dietary lipid spares the protein to perform its function i.e. build up of tissues (Parazo, 1996). However, a high level of energy may produce fatty fish and decrease food consumption (Page and Andrews, 1973; Prather and Lovell, 1973). By providing same protein to energy ratio in the experimental diet this condition was ruled out in the present study.

In fishes, next to nutrient composition of diet the important extrinsic factor that affects the growth performance and nutrient utilization in brackishwater environment is rearing salinity. Mullets, which are cultured in coastal ponds, were also exposed to wide changes in salinity. Studies on salinity effects on growth and feed conversion efficiencies are almost unknown for many euryhaline species (Canagarathnam, 1960; Brett, 1979). The results of the present study indicate that the salinity has significant influence on percent growth increase, specific growth rate, food conversion efficiency and food conversion ratio in *M. cephalus*. The maximum percent growth increase of 77.80 ± 3.34 and 70.52 ± 2.89%, specific growth rate of 2.740 ± 0.11 and 2.573 ± 0.11%, food conversion efficiency of 31.05 ± 1.27 and 29.40 ± 1.26% and food conversion ratio of 3.21 ± 0.13 and 3.40 ± 0.14 recorded for those fish fed on 40% protein diet with carbohydrate (20%) and lipid (15%) at 15ppt salinity indicated their preference in intermediate salinity levels than the low (5ppt) and high salinity (30ppt). This is in consistence with the findings of Paulraj and Kiron (1988) for *Liza parsia*. They reported that, the survival and food conversion in *L. parsia*, fry was high in salinity levels ranging from 5 to 25ppt and very low for those reared at 30 to 35ppt salinity. Desilva and Perera (1985) have also reported that of the four salinities tested
(0, 5, 10 and 15 ppt) for young *Tilapia nilotica*, the salinity level of 10 ppt was optimum for maximum growth. Rao (1974) has also made a similar conclusion and found out that the salinity range of 5 to 14 ppt was optimum for early development of *Fundulus pervipinnis* i.e., for embryonic development, maximum yolk conversion efficiency, largest larval size and maximum viable hatch in the eggs and larvae.

Harel *et al.* (1998) reported that salinity within the range of 25 to 40 ppt had a profound effect on grey mullet *M. cephalus* larvae and they suggest that the grey mullet should be reared at low salinities from egg incubation to the time at which larvae develop a functional swim bladder. After that to accelerate their growth rate, grey mullet should be cultured in full sea water. Similar findings of high growth rate with lower food conversion ratios in sea water as compared to brackish or fresh water were also demonstrated in juvenile red tilapia hybrids (Watanabe *et al.*, 1988; Ernst *et al.*, 1989; Clark *et al.*, 1990).

The effects of different levels of salinity (30, 20, 10 and < 1 ppt) on the growth, food intake and food conversion efficiency of young grey mullet (*M. cephalus*) were studied by Desilva and Perera (1984) and they reported that the maximum growth efficiency was found to occur in 15ppt salinity. The percentage conversion efficiency of fish fed on an excess diet at 10ppt was the highest and when a constant ration was given the percentage conversion efficiency was found to decrease with increase in salinity. Chervinski (1988) reported that when fingerling *Trachinotus ovatus* were subjected to abrupt and gradual changes from sea water salinity to various salinity, the growth rate was better in 10ppt salinity than from other high and low salinities.
Growth is significantly reduced in fishes, including euryhaline species, following exposure to increased salinities (Mckay and Gjerde, 1985; Usher et al., 1991). The negative effects of saline conditions on growth of juvenile fish are generally manifested as a decrease in food intake, food absorption and conversion efficiencies, and in biochemical processes that control digestion and nutrient absorption by the gut (Macleod, 1977; Usher et al., 1990, 1991). In big head carp fry, growth was high between 0 and 2ppt particularly during 3rd and 4th weeks of rearing and this suggests that their conditions are iso-osmotic relative to the fish internal environment, a condition where the energy for maintaining homeostasis becomes minimal. Exposure of bighead carp fry to 4ppt and beyond, shift energy normally intended for tissue assembly and repair to osmoregulation, resulting in decreased growth (Febry and Lutz, 1987). The best growth performances exhibited by *M. cephalus* at 15ppt salinity in the present study may be attributed to the iso-osmotic conditions prevailing in that particular salinity related to internal environment.

The nutrient digestibility and nutrient retention efficiency of *M. cephalus* recorded in the present study was also maximum for those fish reared at 15 ppt salinity and above and below this level the values were less. Despite the high osmoregulatory capability of juvenile mullet, the above results indicate that salinity has a significant effect on nutrient metabolism of this species. At low and high salinities of 5 and 30ppt, which are, hypo / hyper osmotic to blood plasma significantly impaired the digestion and nutrient retention.

In a study on the osmoregulatory capacity of spotted grunter, Bussiahn (1992) demonstrated the iso-osmotic concentration to be 12ppt. The present study also illustrated the well-developed euryhaline character of this species since blood
osmolarity varied by less than 5% over a test range of 5 to 30ppt. The limits of salinity tolerance are defined by the concentrations where a constant blood plasma osmolarity is no longer maintained (Martin, 1990). Disturbance of the plasma osmolarity is indicative of disruption of the \( \text{Na}^+ \) and \( \text{K}^+ \) homeostatic balance which proceeds metabolism disfunction and ultimately results in poor growth performances and nutrient metabolism and even cause mortality (Woo and Fung, 1981). By definition, euryhaline fish are able to maintain blood plasma concentrations at a constant level in ambient salinities, above and below the osmotic concentration of blood plasma (Blaber, 1974; MenI, 1974; Martin, 1990). However, Nordlie (1978) noted that extremely euryhaline fish such as \textit{Oncorhynchus mykiss} allow the internal concentrations of blood plasma to vary considerably depending on the ambient salinity concentration.

The effect of salinity on the growth of fish often had been attributed to the metabolic cost of osmotic and ionic regulation. The usual hypothesis proposed is that the energetic cost of the ion regulation is the lowest in an iso-osmotic environmental where the ionic gradients between water and blood are minimal and that this energy saving is substantial enough to enhance growth and nutrient retention (Morgan and Iwama, 1991). The present results support this hypothesis. In an iso-osmotic concentrations of 15ppt, the nutrient utilization and nutrient retention efficiencies of juvenile \textit{M. cephalus} were significantly higher than that of those reared at low (5ppt) and high (30ppt) salinities. This finding is consistent with many other studies, which showed enhanced growth performance and nutrient retention in an iso-osmotic environment (Deacon and Hecht, 1999).

Further more, osmoregulation may also have secondary effects which are not directly related to ion exchange between blood and water. For example,
assimilation to a different salinity may cause changes in blood haemoglobin content (Guernsey and Poluhowich 1975; Woo and Fung, 1981) and changes in hormone level (Avella et al., 1990). It has also been hypothesized that modified drinking rates resulting from changes in salinity (Conte, 1969) affect stomach evaluation rate, rate of food movement through the intestine, pH of the gastric fluid, food breakdown and consequently adsorption efficiency (Desilva and Perera, 1976; Mac Leod, 1977; Ferraris et al., 1986). The pattern of food conversion and protein efficiency ratio observed in juvenile spotted grunter did not support this hypothesis since both factors were negatively affected at lower salinity levels (5ppt), but were similar at the iso-osmotic concentration (12ppt) and above 25ppt and 35ppt. Since other marine fish, such as European sea bass, *Dicentrarchus labrax*, also have optimum food conversion efficiencies between isosmotic salinity and 35ppt (Dendrinos and Thorpe, 1985) it appears that absorption and digestion are not compromised by the drinking rate as salinity increases. In the present study too, the nutrient digestibility coefficient of *M. cephalus* did not show any significant variation (P<0.05; “t” test) between rearing salinity at constant dietary protein level.