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Review article on Mullet larval Acclimatization and Determination of their nutritional requirements in Egypt

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ABSTRACT

A review of our work on mullet acclimatization during last decade is given in the present article. The effect of water salinity and starting point of acclimatization between 0.0 and 35 ppt on survival and growth of striped mullet larvae were estimated. High mortality rate was found with the striped mullet maintained at the higher salinities (20 ppt and 35 ppt) and also at the lower salinity (0 ppt). Inclusion of wild larvae at salinity similar to the wild salinity for 24 h in the laboratory conditions and start in acclimatization at the second day was recommended. This may help explain the relative paucity of mullet larvae captured in marine sites of the Mediterranean Sea. Larvae are usually very different from their adult counterparts in their simpler digestive systems, which lead to a limited secretion of the digestive enzymes. To study the effect of heat-treated feed and exogenous zymogen® on survival and growth of grey mullet larvae (*Liza ramada*) (Risso, 1826), three experiments were carried out. It has been found that exogenous enzymes from live prey are important to proteolytic activity in larvae. The exogenous added enzymes are also having the same activities. The action of these enzymes is to digest the food and also to activate the endogenous enzymes by cleaving the inactive zymogen. Treating the diet by heat and pressure concentrate on the aspects to improve product quality and make the best nutritional use of the raw materials. It can also sterilize the product and give the flexibility to utilize a wider range of raw materials while maximizing their nutritional quality. Determination of vitamin requirements are also involved in the present article. Vitamins and mineral mixture as will as vitamin C requirements of striped mullet (*Mugil cephalus*) larvae in glass aquaria were determined. The best gain and survival rate were achieved with 0.9 and 1.1 % of the commercial mixture of vitamins and minerals in the diet. Also the minimum dietary vitamin C requirements for the best growth, survival and feed utilization of striped mullet was 40-mg /kg diet. The economically important factor currently limiting production of mullet in intensive culture system is the absence of diets that maximize growth, while taking into account demands for nutrients and vitamins, which are necessary to maintain health. Data in which various dietary protein levels from 14 to 38% at 245 kcal ME/100g diets, support that striped mullet maintained at 26% dietary crude protein level exhibited superior growth characteristics when lipid content was about 6.3%. In relation to the interaction between dietary ME and protein levels, striped mullet larvae exhibited the greatest survival, weight gain and feed utilization when they were maintained at 22% dietary protein with 250 kcal ME /100g diet (lipid contents about 15%) than the other protein levels at all ME levels used.

Keywords: Acclimatization, nutritional requirements, mullet larvae.

INTRODUCTION

Mullet (*Mugillidae*) are considered as one of the important fish species for mono and polyculture systems in Egypt. They represent some of the most promising species for commercial aquaculture and have strong market demand and high price in many countries (e.g. Brazil, Cuba, Colombia, Venezuela, Spain, Egypt and elsewhere) (Benetti and Fagundes Netto 1991, Guinea and Fermandez, 1991 and El-Dahhar, 2000 a). Mullet inhabit all coastal waters lying between 42° north and 42° south latitudes. They usually move into groups within coastal waters and enter lagoons, estuaries and rivers in order to feed, but spawn in seawater (Nash and Shehadeh 1980 Whitehead *et al.*, 1986). Mullets are also good candidates and play an important role in the fisheries and fish farms of tropical and subtropical countries of the world (Nash and Shehadeh 1980). Wild-caught mullet fish larvae is one of the most important and economical source for aquaculture farming in Egypt, since the artificial spawning is still an expensive way for producing these larvae (Nash and Shehadeh 1980). Increasing demands for fish by consumers have intensified fish farming in several ways of production. However, poor survival rates of the wild mullet larvae is a limiting factor in mullet production and many investigations are needed to determine environmental and nutritional requirements of mullet fish specially in the period of larvae culture (El-Dahhar 1999).

The less developed digestive system of marine fish larvae leads to a limited secretion of the digestive enzymes. Larvae are usually very different to their adult counterparts in their simpler digestive systems (Govoni et al., 1986). However, the development of gastro-intestinal complexity is correlated with change in the digestive enzymes (De Silva and Anderson, 1995). The activities of protein degrading enzymes can be correlated with developmental changes in the gastrointestinal tract associated with the change from the intracellular to intra-gastrointestinal protein (Baragi and Lovell, 1986). Detailed study of digestive tract development of Walford and Lam (1993) confirmed the observations of Lauff and Hofer (1984) indicated that exogenous enzymes from live prey are important to proteolytic activity in larvae. The exogenous artificial zymogens also have the same activities (El-Dahhar, 1999). The action of these enzymes is to digest the food and also to activate the endogenous enzyme by cleaving the inactive zymogen (De Silva and Anderson, 1995).

The poor digestion in larval stages of marine fish resulting from such a primitive digestive system indicated that they must be fed live zooplankton. On the other hand, for rapid growth of marine fish farming any diet that can reduce dependence on live prey production is of great technical and economical interest (Person Le Ruyet et al. 1993). Although, live pray are still the most reliable, artificial diets have been used to cover the nutritional requirements in a cheaper way for intensive larviculture. But, when natural components have been included in the diet, digestive enzymes are absent or inactivated through food manufacture. The incorporation of feed additive to stimulate the larva's digestive secretion may be helpful. Also, supplementation of commercial digestive enzymes can enhance food assimilation (Person Le Ruyet et al. 1993). Kolkovsky et al. (1990) provided that suitable levels are used. Digestion of live or artificial food by exogenous and endogenous enzymes in the digestive tract of larval fish has been studied for several years (Dabrowski and Gogowski, 1977 a and 1977 b). Also, there are many reports about exogenous enzyme addition to larval diets to enhance survival rate and feed efficiency (Dabrowski and Gogowski, 1977 c, Dabrowski et al., 1979, Lauff and Hofer, 1984, Tandler and Kolkovsky 1991).

Economically important factor currently limiting production of mullet in intensive culture system is the absence of diets that maximize growth, while taking into account demands for nutrients such as vitamins which are necessary to maintain health. The vitamin requirements of the majority of fish species have not been determined. As a result, the data obtained on some fish species (eg. salmonids, carp or catfish) are usually applied to other species and most diets formulated on the base of vitamins data published by the United States National Research Council (NRC, 1993). However, few studies have been undertaken to assess the influence of diet composition on immune response, although it has been demonstrated that deficiency and/or excess of vitamins, mineral and other feed components may be of significance (Landolt, 1989).

High mortality of the mullet fish larval species is due to some different factors, such as:

- The physico-chemical properties and sudden change in salinities.
- The stress to which the fish is exposed during transportation.
- The less developed digestive system of marine fish larvae.
- Limited secretion of the digestive enzymes in the larval intestine.
- Exogenous enzymes from live prey are important to proteolytic activity in larvae.
- Insufficient live feed in the target environment and
- The unknown larval feed requirements.

This review article was focused on the above factors including our work to explain how it could be possible to solve the problem of high mortality of mullet fish larvae during the period of acclimatization.

Salinity Acclimatization

El-Dahhar *et al.* (1999) estimated the effect of water salinity (0, 5, 10, 15, 20 and 35 ppt) on survival rate percentage of striped mullet larvae. It was observed that increasing water salinity had a marked effect on survival rate. After 24h, a high mortality rate was reported in the highest and the lowest salinity concentrations (35 and 0 ppt). The higher the salinity, the less was the time spent for the larvae to suffer 100% mortality. The same authors also found that mortality was 100% for all treatments after 60 days except 0.0 and 5.0 ppt treatments having 1.1 and 8.0% survival rate respectively. More time was spent for the larvae to suffer 100% mortality with decreasing water salinity. The experimental fish health was poor; the dead fish suffered from disease syndromes (Figure 1) .

Figure 1: *The dead larvae suffer disease syndromes*.

Some authors indicated that mullet fish prefer brackish water and grow faster in such water areas of the subtropics (Gosline and Brock 1965, Nash and Shehadeh 1980). Pillai (1975) reported that mullet could be stocked in different salinities even in fresh water lakes, but need acclimatization when the salinities are considerably different in capturing and stocking sites. The newly hatched mullet larvae were found to have a limited tolerance for salinity fluctuations, but no data have been reported regarding larval growth in varying salinities during their rearing period (Sylvester *et al*. 1975; Lee and Menu, 1981; Walsh *et al*. 1989, Murashige *et al*.1991). However, Devaneson and Chaco (1943) and El-Dahhar *et al*., (1999) found that survival rate of mullet fry could be increased by gradual salinity acclimatization. El-Dahhar *et al*. (1999) designed an experiment to evaluate the starting point salinity acclimatization at which the larvae could tolerate to start acclimatization. Means of survival rate of the grey mullet larvae, which maintained at five starting points of descending salinity acclimatization (3, 6, 9, 12 and 15 ppt) are presented in Table 1.

After 24 days, survival rates of the larvae started acclimatization at 3 and 12 ppt salinity were higher (65.6 and 70%) significantly ($P<0.05$) than all other treatments. They were still the higher until after 30 days. However, survival rate of the larvae started at 3 ppt salinity and was the highest until the end of the experiment after 54 days. Also, on the average, starting points of acclimatization of 3, 6 and 12 ppt salinity had the best significant ($P<0.05$) effect on survival rate at the end of this experiment. Their survival rates were 33.3, 24.4 and 24.4 % respectively. At the conclusion of this work it was found that starting acclimatization at salinity similar to the natural salinity from which the larvae were caught (about 7 ppt) was the best starting point for the wild mullet larval acclimatization at laboratory conditions.

Table 1: *Means of three replicates of survival rate percentage of grey mullet larvae maintained at the five descending starting points of salinity acclimation (3, 6, 9, 12 and 15 ppt).*

	Starting point salinity acclimation ppt				
Days No	3	6	9	12	15
Start	100	100	100	100	100
6	75.6 ^{A}	60.0 ^{AB}	46.7 ^{B}	72.2 ^{A}	71.0 ^{A}
12	67.8 ^{A}	57.8 ^{AB}	45.6 ^{B}	72.2 ^{A}	55.6 ^{AB}
18	67.8 ^{A}	56.7 ^{AB}	45.6 ^{B}	71.1 ^{A}	51.1 ^{AB}
24	65.6 ^{A}	54.4 ^{AB}	42.2 ^{B}	70.0 ^{A}	43.3 ^{B}
30	65.6 ^{A}	45.6 ^{B}	33.3 ^{B}	56.7 ^{AB}	37.8 ^{B}
36	57.8 ^{A}	41.1 ^{B}	23.3 ^{C}	41.1 ^{B}	31.1 ^{C}
42	53.3 ^{A}	36.7 ^{B}	13.3 ^{C}	33.3 ^{B}	28.9 ^{B}
48	44.4 ^{A}	30.0 ^{B}	5.6 ^{C}	26.7 ^{B}	26.7 ^{B}
54	33.3 ^{A}	24.4 ^{B}	3.3 ^{D}	24.4 ^{B}	13.3 ^{C}

Numbers within the same row followed by different superscript letters are significantly different ($P < 0.05$). (El-Dahhar et al., 1999)

Feed Treatments And Additives

1 Heat And Zymogen Treatments Of Feed

Treating the diet by heat and pressure concentrate on expecting to improve product quality and make the best nutritional use of the raw materials. It can also sterilize the product and give the flexibility to utilize a wider range of raw materials while maximizing their nutritional quality (Botting, 1991; De Silva and Anderson, 1995).

The ingested food in the larval stage is passed directly into the intestine because the stomach is not yet developed and the gastric glands are not fully developed (Tanaka 1969; Govoni, 1980 and O'Connell, 1981).

They also stated that the digestive system is generally considered immature when compared with that of the adult. In the intestinal lumen, the food is first digested by the pancreatic enzymes, then digested by the enzymes of the intestinal brush border, and absorbed by the intestinal epithelial cells (Dabrowski *et al.*, 2003). However, the larval intestine is not able to digest all ingested nutrients. In order to increase the mullet larval capability to utilize heat -treated artificial diets, the effect of adding different levels of exogenous zymogen® was investigated (El-Dahhar, 1999). The final aim of this work was to asses the significance of the heat-treated and exogenous zymogen® supplemented diets on survival and growth of grey mullet larvae collected from the wild to decrease dependence on live food. In this trial, we used six experimental test diets either heat treated in an autoclave using a maximum pressure of 1.2 Kg / cm^2 for 15 min or supplemented with exogenous zymogen® at the rates of 0, 2, 4, 6 or 8 % of the diet (El-Dahhar, 1999). From this trial, the survival rate percent of the larvae maintained on the heat treated diet and diet with 4% zymogen® were significantly higher as compared to the larvae maintained on the other diets starting from day 7 to day 10. The same trend was also observed until the end of this preliminary trial. In general, the quality of the experimental fish was poor; the dead fish had skeleton abnormalities when compared to the normal larvae (Figure 2).

From this point of view, in the same research we used four heat treating duration (0, 10, 20 or 30 min) with the same previous pressure and three exogenous zymogen® supplementation rate (0, 2 or 4 %) in a 4X3 factorial arrangement to evaluate their effects on survival and growth of grey mullet using a balanced diet. The treatment was repeated in three glass aquaria (Table 2).

Table (2) shows that percentage of survival rate increased significantly (P< 0.01) with increasing zymogen® inclusion in the diet starting from week 2. However, heat treatment of the diet had a significant effect (P< 0.05) on larval survival rate starting from week 4.

A 57.9% survival rate for larvae offered diet with 4% zymogen® was significantly (P< 0.01) higher than 45.0 and 43.3% for larvae offered the diet with 0 and 2% zymogen® respectively after 6 weeks treatments. Heat treating time of the diet had a marked effect on the final biomass of grey mullet after four weeks of the second experiment. Weight gain (percent weight gain) and final biomass showed a general increase with increasing exogenous zymogen® inclusion in the diet. Larvae maintained at diet without zymogen® inclusion gained 11.15% on the average which was significantly (P< 0.01) less than 16.93% and less than 26.19% on the average observed for larvae offered diet with 2 and 4% zymogen® inclusion respectively (Fig. 3). Also, heat-treating time had a significant effect (P<0.01) on percent weight gain of grey mullet larvae maintained for six weeks on the treatment of this experiment. However, heat treatment had a significant (P<0.05) effect on survival rate. 20-min heat treatment for diet showed a high survival rate for grey mullet larvae (55.6%) as compared to 49.4, 46.1 and 43.9% observed for larvae offered heat-treated diet for 30, 0 and 10 min respectively.

A significant (P<0.01) interaction between the two factors (heat treatment and exogenous zymogen®) was also observed on percent survival rate starting after week 1 until the end of the experiment. No interaction between the factors was observed in the criteria of growth.

2. Vitamins Treatments

a. Vitamins and mineral mixture

The commercial mixture of vitamins and minerals was examined in an experiment. We used seven concentrations (0, 0.1, 0.3, 0.5, 0.7, 0.9 and 1.1 %) to determine its effect on survival and growth of striped mullet (El-Sayad, Rania, 2001). Gain and survival rate of striped mullet (0.1g initial body weight) increased with increasing the vitamin and mineral mixture concentration in the diet. The best significant weight gain and survival rate were achieved with 0.9 and 1.1 % of the commercial mixture of vitamins and minerals in the diet (P< 0.05). Their final body weights were 0.54 and 0.60 g compared to 0.18 g for level 0.7% commercial vitamin mixture. They also achieved 77.7, 77.7 and 31.1 % survival rate respectively.

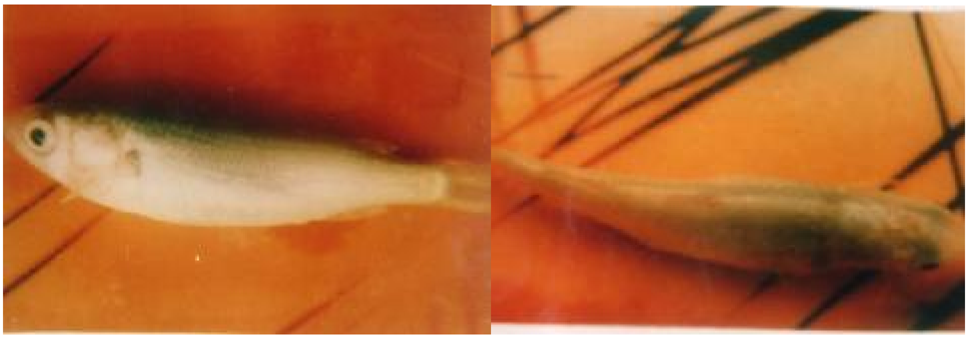

Figure 2. *Left, apparently normal mullet larvae, Right, abnormal shape of dead larvae indicating deformed skeleton*

Table 2: *Weekly survival rate % of grey mullet offered the test diets over a 6-wk growth trial in the second experiment.*

Zymogen® Level %	Heat treating time (Min)	Week 1	Week 2	Week 3	Week 4	Week 5	Week 6
0	0	81.7	75.0	75.0	61.7	65.7	53.4
	10	60.0	58.3	56.7	46.7	36.7	30.0
	20	75.0	73.4	73.4	63.4	55.0	55.0
	30	75.0	75.0	71.7	51.7	43.4	41.7
	Mean±SE	72.9^{a}±2.8	70.4^{c}±2.4	69.2^{b}±2.4	55.8^{b}±2.4	47.9^{b}±2.8	45.0^{b}±3.3
2	0	85.0	81.7	75.0	58.3	41.7	28.4
	10	66.7	65.0	65.0	51.7	48.4	45.0
	20	76.7	75.0	73.7	65.0	59.8	55.0
	30	81.7	80.0	76.7	56.7	48.4	45.0
	Mean±SE	77.5^{a}±4.5	75.4^{b}±2.7	72.6^{b} ± 2.2	57.9^{b}±1.7	49.5^{b}±2.5	43.3^{b}±3.2
4	0	66.7	66.7	66.7	63.4	58.4	56.7
	10	86.7	86.7	83.4	70.0	60.0	56.7
	20	88.4	86.7	83.4	75.0	65.0	56.7
	30	85.0	83.4	83.4	75.0	68.4	61.7
	Mean±SE	81.7^{a}±3.8	80.8^{a}±3.2	79.1^{a}± 3.0	70.8^{a}±1.9	62.9^{a}±2.5	57.9^{a}±2.1
Mean±SE	0	77.8^{a}±2.9	74.4^{a}±3.0	72.2^{a}± 2.1	61.1^{b}±1.6	52.2^{bc}±3.3	46.1^{bc}±4.4
	10	71.1^{a}±2.9	70.0^{a}±4.6	68.3^{a}± 4.1	56.1^{c}±3.9	48.3^{c}±3.9	43.9^{c}± .6
	20	80.0^{a}±4.5	78.3^{a}±2.7	76.8^{a}± 2.7	67.8^{a}±2.3	59.9^{a}±2.6	55.6^{a}±1.9
	30	80.6^{a}±3.8	79.5^{a}±2.9	77.2^{a}± 3.2	61.1^{b}±3.7	53.4^{b}±4.1	49.4^{b}±3.4

Numbers are the average of three replicates. Means within the same column with different superscript are significantly different. (El-Dahhar, 1999)

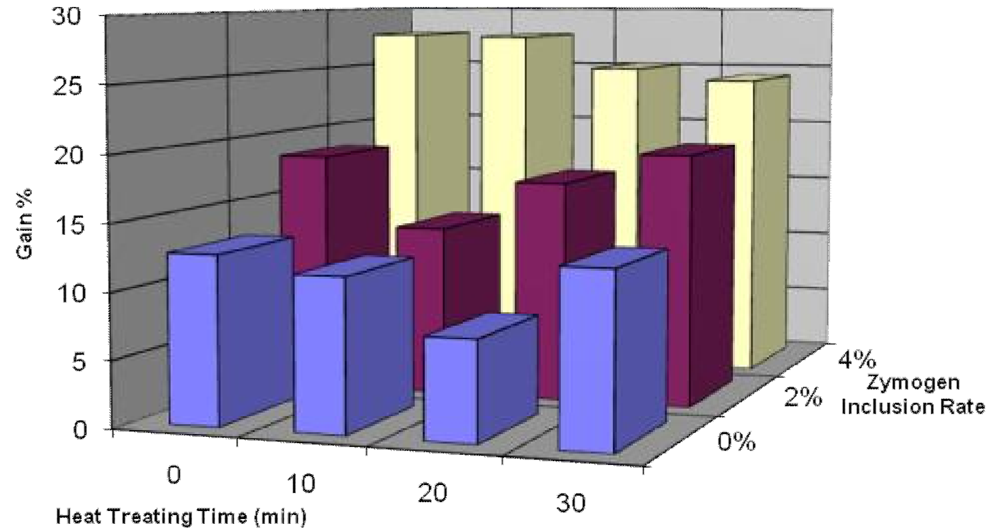

Fig. 3: Growth results of gray mullet larvae fed 3 levels of exogenous zymogen each in heat treated diets for 4 periods of time, at the treatments of the second experiment (El-Dahhar, 1999).

b. Ascorbic acid , Vitamin C

Ascorbic acid is a water-soluble vitamin synthesized from glucuronic acid by some animals, but important cultured fish species are unable to synthesize it (NRC 1993). Generally, feed ingredients do not contain enough ascorbic acid to meet the dietary requirements of fish, so it is necessary to supplement this vitamin in aquaculture diets (Masumoto *et al*. 1991).

In well-established aquaculture species such as channel catfish *Ictalurus punctatus*, and hybrid striped bass (*Morone chrysops* × *M. saxatilis)* dietary vitamin C requirements have been widely studied. The minimum dietary vitamin C requirement for normal growth and best weight gain has been determined to range from 25 to 50 mg/kg diet for channel catfish *Ictalurus punctatus* (NRC 1993) and to be 22 (± 6) mg/kg for hybrid striped bass *Morone chrysops* × *M. saxatilis* (Sealey and Gatlin 1999). Based on the concentrations of vitamin C in the liver and kidney as an indicator for dietary ascorbic acid requirements, Blom and Dabrowski (2000) recommended a level of 360 mg/kg diet as the dietary vitamin C supplementation level necessary to maintain tissue stores of the angelfish *Pterophylum scalare*. Generally, vitamin requirements of fish increased when they are grown in intensive culture systems (Sandnes 1991).

Using the best result of the vitamin and mineral mixture on survival and growth of stripped mullet achieved in the previous work of El-Sayad, Rania (2001), another experiment was conducted to asses the effect of vitamin C on the survival and growth of the mullet larvae during the period of acclimatization (El-Dahhar, 2000 a). We used seven ascorbic acid concentrations 0.0, 10, 20, 30, 40, 50 and 60 mg / kg diet to feed 0.2 g initial BW striped mullet larvae for six weeks. The data of the experiment of vitamin C are shown in Table 3. In this work, El-Dahhar, (2000 a) demonstrated that striped mullet larvae have dietary vitamin C requirements. Fish fed the diet with 0.0, 10 and 20-mg vitamin C / kg had reduced weight gain and feed efficiency. Reduced weight gain had been shown to be due to vitamin C deficiency signs for many fish species (Hilton *et al*. 1978; Sandnes 1991; Li *et al*. 1998; Sealey and Gatlin 1999). Fish fed these deficient diets in this study exhibited poor survival and abnormalities in skeleton. High mortality and gross deficiency signs in fish maintained on deficient vitamin C diets have been reported for many fish species (Chávez de Martínez 1990; Chávez de Martínez and Richards 1991; Shiau and Jan 1992).

Structural deformities due to improper cartilage formation such as lordosis and scoliosis were observed Figure 4 (El-Dahhar, 2000 a). They have been observed rottenly in fish fed vitamin C deficient diets (Wilson and Poe 1973; Lim and Lovell 1978; Sealey and Gatlin 1999). Masumoto *et al*. 1991 reported that collagen is a major component of the body skeleton, which is necessary for normal growth, and ascorbic acid has an important role in its formation. It has been shown that ascorbic acid is rapidly taken up from the areas where collagen is formed, namely the skin, caudal fin and in the heavy cartilage of the snout, head and jaw, gill support cartilage and bones (Halver, 1988). El-Dahhar, (2000 a) also reported that striped mullet fed supplemental dietary vitamin C above 30-mg/kg had improved weight gain, specific growth rate (SGR), feed efficiency, survival and no observed growth abnormalities. Supplemental vitamin C levels above 40-mg/kg diet resulted in no substantial additive effect on weight gain, survival and feed utilization (protein efficiency ratio, PER, protein productive value, PPV and energy retention, ER). Sato *et al*. (1982) used bone and skin hydroxyproline to proline ratio as an indicator for ascorbic acid status of rainbow trout. Fish receiving ascorbic acid below 50 ppm had depressed ability to synthesize normal collagen evidenced by a lower ratio. Fish receiving above 100 ppm showed a higher and constant ratio, indicating restored collagen formation. In the study of El-Dahhar, (2000 a), increasing dietary vitamin C, improved feed efficiency and survival of striped mullet fed above 30-mg vitamin C/kg diet which reflect the ability of the vitamin to reduce physiological stress. Since vitamin C have a role in the synthesis of corticosteroids (Sauberlich, 1984), high levels of vitamin C has been proposed as being beneficial for reducing the effects of physiological stress in fish (Jaffa, 1989; Hardie *et al*., 1991).

3. Determination of Mullet Feed Requirements

a. Maintenance and maximum growth requirements of protein and energy

Mullet growth like most fish species depends to a large extent on supplementary food, and the cost of feed constitutes the principal part of the total production costs. Furthermore, feeding rate for mullet is still based on the feeding rate of other species rather than on mullet data. Determination of feeding rate with respect to the nutritional requirements, dietary composition and the effect of environmental conditions may help in saving feed or promoting fish growth. For establishing feeding level of a fish species, some major factors should be taken into consideration e.g. maintenance requirements, growth requirements and feed utilization for all purpose (Hepher *et al*., 1983). Mullet maintenance requirements were determined in several studies.

Figure 4: *Structural deformities due to improper cartilage formation left: lordosis and scoliosis right. (El-Dahhar, 2000 a).*

Table 3: *Mean weight gain, percent survival, feed intake and feed conversion ratio of striped mullet (0.2g initial body weight) fed various levels of vitamin C for 6 wk. Values represent mean of N = 3 determinations / treatment. Means (± SE) in the same column sharing the same letter are not significantly different (P<0.01).*

Diet Number	Vitamin C level mg / kg	Final Weight (g)	Weight Gain (g)	Survival %	Dry mater feed intake g / fish	Feed conversion ratio *
1	0.0	0.4 ± 0.002 c	0.23 ± 0.01 c	21.7 ± 1.7b	1.0 ± 0.01 c	4.3 ± 0.1 a
2	10.0	0.5 ± 0.003 c	0.25 ± 0.01 c	20.0± 1.1 b	1.0 ±0.0 bc	4.0 ±0.1 ab
3	20.0	0.5 ± 0.01 bc	0.3 ± 0.01 bc	38.3 ±1.7 b	1.0 ±0.0 bc	3.7±0.1abc
4	30.0	0.6± 0.02abc	0.4± 0.02abc	90.0± 1.1 a	1.1±0.00ab	2.8±0.1bcd
5	40.0	0.65±0.01ab	0.45±0.01ab	93.3±1.7 a	1.13±0.01a	2.5±0.1 cd
6	50.0	0.68 ± 0.01a	0.48 ± 0.01a	95.0±1.13a	1.13±0.1a	2.4± 0.1cd
7	60.0	0.68 ± 0.01a	0.48 ± 0.01a	95.0 ± 1.1a	1.1±0.01ab	2.3± 0.04 d

** Feed conversion ratio = Dry feed fed (g)/wet weight gain (g). (El-Dahhar, 2000 a)*

In a feeding trial using different feeding rates 1, 3, 5, 7, 9, 11 and 13 % of BW striped mullet (*Mugil cephalus*) (0.195-g initial BW) were fed a diet containing 38% crude protein for 6 weeks to determine protein and energy requirements for maintenance and maximum growth (Table 4) (El-Dahhar, 2000 b). Weight gain in a plot against protein or energy intake (Fig. 5 and 6) indicate that daily intake of 34.6 mg protein intake and 345.6 cal gross energy intake/g BW daily and more got the maximum growth of striped mullet. The relationships between BW gain (Y) and the increase of protein and energy intake (X), could be expressed by the equations Y = - 58.06 + 16.20 X for protein (Fig. 5) and Y = - 57.98+ 1.62 X for gross energy (Fig. 6).

Maintenance requirements calculated from these equations (as BW gain = zero) of striped mullet larvae daily were found to be 3.6 mg protein and 35.8 cal gross energy/g BW daily. Also, the increase in body protein and energy (Y) with the increase in protein and energy intake (X) could be expressed by the equations Y = - 11.33 + 2.68 X; r^2 = 0.98 for protein (Fig. 5) and Y = - 85.67 + 3.48 X; r^2 = 0.99 for energy (Fig. 6).

From the regression analysis, maintenance requirements could be calculated at Y = zero. 4.2 mg protein and 24.6 cal gross energy were the maintenance requirements of striped mullet (0.195g BW) /g of the fish BW daily. From these mathematical models, maintenance requirements of striped mullet (0.195-g initial BW) were established to be in the range of 3.6 - 4.2 mg protein / (g BW .d) and 24.6 - 35.8 cal gross energy /(g BW .d). Thus the author recommended 3.9 mg protein /g BW and 30.2 cal gross energy /g BW daily as maintenance requirements for 0.195g striped mullet fry. The author also recommend 26% dietary protein to be optimum level for striped mullet 0.2-g for maximum growth and feed utilization. This protein optimum is similar to that reported for grey mullet (*Mugil capito*) of weight 0.8 and 3g by Vallet *et al*. (1970) and for grey mullet 2.5g by Papaparaskeva-Papoutsoglou and Alexis (1986).

Most fish studied are either carnivorous or omnivorous. Mullet have been variously described as vegetarian, planktophagous, detritivorous, omnivorous and even carnivorous (Brusle, 1981; Benetti and Netto 1991). They change their food performances from carnivorous to herbivorous as they grow bigger (Albertini-Berhaut, 1974). Thus their ability to use protein as an energy source could be affected by dietary crude protein levels (Alexis and Papaparaskeva-Papoutsoglou, 1986). The same authors also stated that the metabolic behavior of the fish studied so far to digest protein could be attributed to various factors. Genetically utilizing protein as an energy source is an important factor, so the metabolism of carnivorous fish might geared towards the use of protein as an energy source which affected by dietary protein level. Dietary energy level is another factor. Doubling the lipid content of the diet for rainbow trout with constant protein level, decreases protein metabolism in the form of glutamic pyrovic transaminase activity in the liver by almost four times (De la Higuera *et al.*, 1977). Lipids are known to be efficiently utilized as an energy source and their protein sparing action has been reported (Lee and Putnam, 1973; Watanabe *et al.*, 1983).

Feed conversion ratio (FCR) as a function of feeding rates is important in the fish culture. Too much rates of feeding, waste feed, approach the rate required for maintenance, decrease fish growth, and increase the rate of conversion. Generally, it was found that when fish were fed at a rate lower than satiation, fish fed the higher protein diets grow faster and hence protein requirements increase. Clark *et al*. (1990) reported a maximum growth rate for Florida red tilapia (*O. urolepis hornorum* X *O. mossambicus* monosex) fed all they can eat in floating marine cages when the diet containing 32% crude protein. 25.5% was the optimum dietary protein level reported for *M. capito* 2.5-g when reared at the laboratory in 150-l glass aquaria with 12 ppt salinity (Papaparaskeva-Papoutsoglou and Alexis, 1986).

b. Dietary protein to energy balance for mullet.

Mullet exhibit numerous traits considered beneficial for aquaculture (Nash and Shehadeh 1980; Papaparaskeva-Papoutsoglou and Alexis, 1986 and El-Dahhar, 2000 b) including the ability to adapt to rear in the polyculture system. The cost of the controlled production of fish depends mainly on the cost of feed, which constitute most of the economical expenses, and crude protein is typically the most expensive component in artificial diets for fish. The optimum dietary protein needed for maximum growth and feed utilization is essential in the formation of well-balanced and low cost artificial diets.

Table 4: *Mean weight gain, feed intake, feed conversion ratio, percent survival, and specific growth rate (SGR) of striped mullet fry (0.2g initial BW) fed at various feeding rates. (El-Dahhar, 2000 b).*

Feeding rate	Protein intake	Gross Energy intake	Weight gain	Feed consumption	Feed conversion ratio	Survival	SGR
(% of BW)	Mg /g BW	cal /g BW	(g/fish)	(g/fish)	FCR [1]	(%)	(% / d)
1	3.85	38.4	0.02±0.01^{C}	0.07±0.0^{C}	3.6±0.5^{A}	18.3±1.7^{C}	0.3±0.04^{C}
3	11.54	115.2	0.13±0.01^{C}	0.24±0.0^{C}	1.9±0.02^{C}	40.0±2.9^{BC}	1.4±0.01^{BC}
5	19.23	192.0	0.24±0.01^{BC}	0.41±0.00^{BC}	1.7±0.02^{C}	63.3±6.0^{B}	2.3±0.02^{BC}
7	26.92	268.8	0.36±0.01^{BC}	0.69±0.01^{BC}	1.9±0.03^{C}	73.3±4.4^{AB}	3.0±0.02^{B}
9	34.61	345.6	0.52±0.01^{AB}	1.09±0.01^{AB}	2.1±0.04^{BC}	85.0±2.9^{A}	3.7±0.05^{AB}
11	42.31	422.4	0.61±0.01^{A}	1.47±0.01^{AB}	2.6±0.06^{AB}	81.7±4.4^{A}	3.9±0.02^{A}
13	45.00	499.2	0.61±0.01^{A}	1.89±0.02^{A}	3.2±0.12^{AB}	85.0±2.9^{A}	3.9±0.02^{A}

Values represent means of N = 3 replicates / treatment. Means (±SE) in the same column having the same superscript are significantly different (P < 0.01).

1. FCR = feed / gain

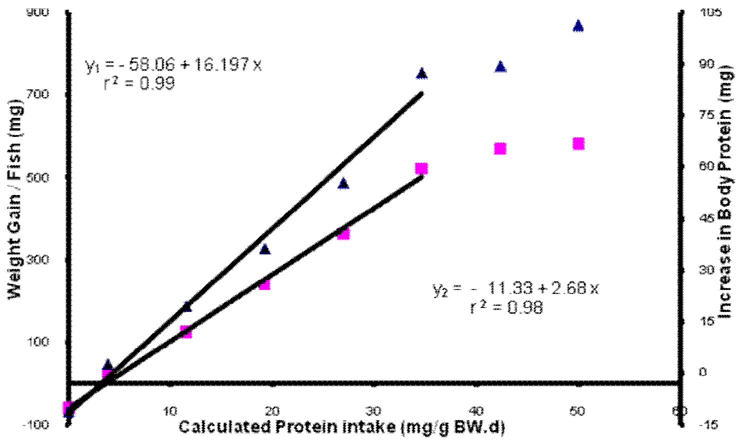

Fig. (5): Effect of increasing levels of calculated protein intake (mg / g BW.d) on striped mullet weight gain and change in body protein (mg). Each symbol represents the mean of three observations.

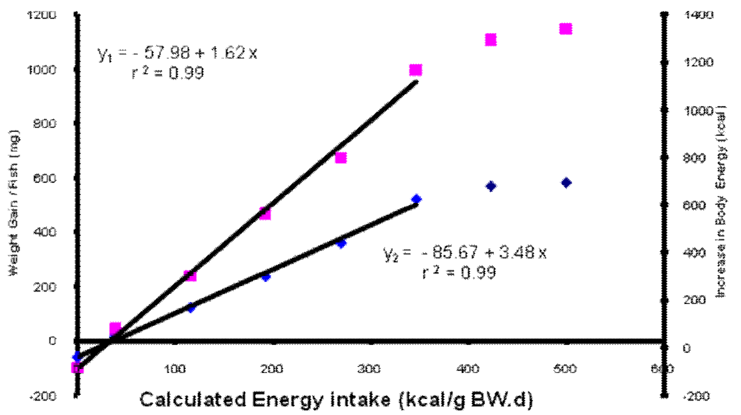

Fig. (6): Effect of increasing levels of calculated energy intake (kcal/g BW .d) on striped mullet weight gain and change in body energy (kcal). Each symbol represents the mean of three observations.

Much of the nutritional research with fish species has focused on minimizing crude protein in the diet for mullet (Albertini-Berhaut, 1974; Papaparaskeva-Papoutsoglou and Alexis, 1986; El-Dahhar, 2000 b and c). These researches tended to reduce the cost of feed. The initial results for striped mullet (*Mugil cephalus*) larvae of 0.2g initial BW fed at incremental dietary crude protein levels 14, 18, 22, 26, 30, 34 and 38% to determine protein requirements, El-Dahhar (2000 b) found that 26% dietary crude protein level is the level needed for maximum growth and feed utilization (Table 5).

Also, Papaparaskeva-Papoutsoglou and Alexis, (1986) fed young grey mullet (*M. capito*), of average initial BW 2.5g, five semi-purified diets containing 12–60% protein. They found that 24% protein was required for maximum growth at 23 ^{o}C. While, Alexis and Papaparaskeva-Papoutsoglou (1986) found that the dietary protein content of 15% resulted in sufficient growth equal to 26, 37 and 50% dietary crude protein levels for grey mullet of 12.5g initial BW.

The optimum protein level of the diet can be lowered if the energy level is increased, due to protein sparing action of energy nutrients (Lee and Putnam, 1973; Machiels and Henken, 1985; Serrano ***et al.***, 1992; Shiau and Peng, 1993; Jantrarotai ***et al.***, 1998 and El-Dahhar, 2000 c). However, increasing energy in catfish diet may decrease feed efficiency and leads to increased fat deposition (Reis ***et al.***, 1989). But, increasing energy in a diet for ***Mugil capito***, indicates a better utilization of protein (Alexis and Papaparaskeva-Papoutsoglou, 1986 and El-Dahhar, 2000 c).

The energy available to mullet by the diet under test is an important factor for the different behavior of fish to use protein as an energy source (Alexis and Papaparaskeva-Papoutsoglou, 1986). Also,

Table 5: *Mean weight gain, feed intake, feed conversion ratio, percent survival, and specific growth rate (SGR) of striped mullet fry (0.2g initial BW) fed at various dietary crude protein levels. (El-Dahhar, 2000 b).*

Dietary protein level %	Weight gain (g/fish)	Feed consumption (g/fish)	Feed conversion ratio FCR [1]	Survival (%)	SGR (% / d)
14	0.2±0.01^{C}	0.9±0.01^{C}	4.06±0.1^{A}	56.7 ± 6.0^{B}	1.82±0.03^{B}
18	0.4±0.02^{C}	1.1±0.03 ^{C}	2.8±0.12^{B}	51.7± 4.4^{B}	2.57±0.1^{AB}
22	0.5±0.02^{BC}	1.2±0.03^{BC}	2.3±0.05^{B}	63.3±6.0^{AB}	3.0±0.1^{A}
26	0.7±0.01^{AB}	1.4±0.02^{AB}	2.1±0.04^{B}	70.0±2.9^{AB}	3.5±0.1^{A}
30	0.7±0.01^{A}	1.5±0.01^{AB}	2.1±0.04^{B}	86.7±4.4^{A}	3.6±0.1^{A}
34	0.7±0.01^{A}	1.6±0.01^{AB}	2.3±0.05^{B}	88.3±1.7^{A}	3.6±0.1^{A}
38	0.7±0.01^{A}	1.7±0.02^{A}	2.34±0.0^{B}	88.3±1.7^{A}	3.6±0.1^{A}

Values represent means of N = 3 replicates / treatment. Means (±SE) in the same column having the same superscript are significantly different (P < 0.01).

1. FCR = feed / gain

De la Higuera *et al*. (1977) indicated that doubling the amount of lipid supplied in rainbow trout diets with a constant protein level, decreased protein metabolism in form of glutamic pyrovic transaminase activity in the liver almost by four times. Lipids are good sources efficiently utilized for energy and have a sparing action for protein to be used as a source of energy. That has been reported for rainbow trout (Lee and Putnam, 1973; Watanabe, 1977 and Watanabe *et al*., 1979), for halibut (Helland and Gisdale-Helland, 1998) and for mullet (Alexis and Papaparaskeva-Papoutsoglou, 1986 and El-Dahhar, 2000 c). This protein sparing action of lipids has also been repeatedly proven (Jürss, 1981). He observed a decrease in transaminase activity with the decrease of protein content and increase in the lipid content of the diet for rainbow trout. Data presented by El-Dahhar, (2000 c) clearly demonstrated that performance of striped mullet (***Mugil cephalus***) maintained at diets containing 175 to 265 kcal ME /100g was superior at 250 kcal ME/100g compared to other treatments when dietary protein level was stable at 26% (Table 6). In this work, El-Dahhar, (2000 c) also stated that striped mullet fed at three levels of dietary protein levels (18% 22% and 26%) and three levels of ME (200, 225 and 250 kcal / 100g diet), weight gain, survival and feed efficiency improved as dietary energy increased up to 250 kcal / 100g diet (Table 7). Differences in performance among protein levels in this trial were most pronounced in fish fed 26% protein diets, which resulted in the fastest growth. In relation to the interaction between dietary ME and protein levels, striped mullet larvae exhibited the greatest survival, weight gain and feed utilization when they were maintained at 22% dietary protein with 250 kcal ME /100g diet than the other protein levels at all ME levels used. These differences in survival and weight gain can be attributed to the increased feed intake and feed efficiency exhibited at 250 kcal ME /100g diets (Table 7). Data from previous study in which various dietary protein levels from 14 to 38% at 245 kcal ME / 100g diets (Table 5), (El-Dahhar, 2000 b), support that striped mullet maintained at 26% dietary crude protein level exhibited superior growth characteristics when lipid contents was about 6.3%. In El-Dahhar, (2000 c) study, increasing the amount of energy in the form of lipid (about 15%) decreased dietary crude protein needed for maximum survival and growth from 26% to 22% (Table 7).

With *Clarias gariepinus* catfish, Machiels and Henken (1985) related growth and feed efficiency to be functions of dietary protein intake. They found that increasing protein intake of fish fed diets with different protein levels improved growth and feed efficiency. In El-Dahhar, (2000 c) study, the data clearly indicated that fish fed the 26% protein diets consumed more protein than those fed lower protein diets and therefore grew and utilized feed better (Table 6). Energy retention (ER), which was low in fish fed the low protein diets is in agreement with that reported for ***M. capito*** (Alexis and Papaparaskeva-Papoutsoglou, 1986).

However, for *M. cephalus*, the PPV was also low for the fish fed the lower (14%) protein diet leading El-Dahhar (2000 b) to conclude that some protein was used for energy. This is in contrast to his finding (El-Dahhar, 2000 c) where PPV of fish fed the low protein diets was similar to that for fish fed higher protein diets. This is probably because fish fed the low protein diets consumed enough non-protein energy so that the energy intake of fish in this group was the highest, known from it's least ER value.

El-Dahhar, (2000 c) found that increasing energy levels over 235 kcal ME/100g had no beneficial effects on fish performance. This agrees with results for *Clarias gariepinus* catfish (Degani *et al*., 1989). However, studies with *C. gariepinus* (Machiels and Henken, 1985), channel catfish *Ictalurus punctatus* (Garling and Wilson, 1976), Nile tilapia *Oreochromis niloticus* (El-Sayed and Teshima, 1992), Mozambique tilapia *O. mossambicus* (El-Dahhar and Lovell, 1995) and rainbow trout *Oncoryhnchus mykiss* (Lee and Putnam, 1973), had shown that increasing both protein and energy to the optimum levels resulted in better growth of fish and improved feed efficiency. The difference may relate to energy source.

Since in studies of Jantrarotai *et al*., (1998) with *Clarias* catfish and El-Dahhar (2000 b) with striped mullet *M. cephalus*, carbohydrate was the major energy source whereas in El-Dahhar's (2000 c) study more lipid were used for energy. The same author tested this assumption, in which corn oil was added in graded levels to achieve three levels of ME. In this work, optimum growth, survival and dietary utilization of striped mullet *M. cephalus* were attained at 22% protein and 250 kcal ME/100g. The protein level was lower and the energy level was the same as the previous result with striped mullet (El-Dahhar, 2000 b). Furthermore, the improved growth rates as well as protein sparing effect were observed in fish fed higher energy diets (El-Dahhar, 2000 c). These findings were not reported in the previous study with striped mullet (El-Dahhar, 2000 b), who found that 26% protein was needed for maximum growth and feed utilization when carbohydrate as a source of energy was higher than lipid. The difference in protein requirement for striped mullet *M. cephalus* (El-Dahhar 2000 c) (22%) and his previous study (El-Dahhar, 2000 b) may be due to the different ratios of carbohydrate and lipid. From these results it could be concluded that striped mullet *M. cephalus* utilize energy with a low ratio of carbohydrate to lipid more efficiently and this resulted in a protein sparing effect. Also, *Clarias* catfish use lipid as a source of energy more efficiently than carbohydrate (Degani *et al*., 1989 and Jantrarotai *et al*., 1998)

Table 6: *Mean final weight, weight gain, feed intake, feed conversion ratio, percent survival, and specific growth rate (SGR) of striped mullet fry (initial wt. = 0.173 ± 0.01) fed at seven dietary energy levels.*

Energy level Kcal ME/100g	Protein level %	Final Weight (g/fish)	Weight Gain (g/fish)	Feed consum-ption (g/fish)	Feed conversion ratio FCR [1]	Survival (%)	SGR (% / d)
175		0.36 ± 0.01 ^{C}	0.19 ± 0.01 ^{C}	1.25 ± 0.02 ^{C}	6.71 ±0.46 ^{A}	34.67 ± 1.3 ^{B}	1.30 ± 0.07 ^{C}
190		0.41 ± 0.01 ^{BC}	0.23 ± 0.01 ^{BC}	1.31 ± 0.04 ^{C}	5.70 ± 0.08 ^{AB}	52.00 ± 2.3 ^{AB}	1.50 ± 0.05 ^{C}
205		0.43 ± 0.01 ^{BC}	0.26 ± 0.01 ^{BC}	1.39 ± 0.01 ^{BC}	5.29 ± 0.12 ^{AB}	61.33 ± 4.8 ^{AB}	1.66 ± 0.02 ^{BC}
220	26	0.61 ± 0.03 ^{BC}	0.43 ± 0.03 ^{BC}	1.69 ± 0.10 ^{BC}	3.90 ± 0.03 ^{BC}	69.33 ± 9.3 ^{A}	2.24 ± 0.08 ^{BC}
235		0.74 ± 0.05 ^{ABC}	0.56 ± 0.05 ^{ABC}	2.11 ± 0.07 ^{BC}	3.74 ± 0.03 ^{BC}	78.67 ± 7.1 ^{A}	2.57 ± 0.04 ^{BC}
250		1.36 ± 0.01 ^{A}	1.18 ± 0.01 ^{A}	3.24 ± 0.06 ^{A}	2.73 ± 0.02 ^{C}	62.67 ± 8.7 ^{AB}	3.66 ± 0.01 ^{A}
265		1.33 ± 0.02 ^{A}	1.16 ± 0.02 ^{A}	3.14 ± 0.02 ^{A}	2.71 ± 0.03 ^{C}	81.33 ± 7.1 ^{A}	3.66 ± 0.03 ^{A}

Values represent means of N = 3 replicates / treatment. Means (±SE) in the same row having the same superscript are significantly different (P < 0.01).

1. FCR = feed / gain.

Table 7: *Mean final weight, weight gain, feed intake, feed conversion ratio, percent survival, and specific growth rate (SGR) of striped mullet fry (initial wt. = 0.329 ± 0.01) fed at three dietary energy levels X three dietary protein levels.*

Energy level Kcal ME/100 g	Protein level %	Final Weight (g/fish)	Weight Gain (g/fish)	Feed consumption (g/fish)	Feed conversion ratio FCR [1]	Survival (%)	SGR (% / d)
	22	0.99 ± 0.02 ^{CD}	0.66 ± 0.02 ^{C}	2.60 ± 0.05 ^{CD}	3.96 ± 0.10 ^{A}	72.0 ± 2.3 ^{D}	1.95 ± 0.06 ^{BC}
	26	1.25 ± 0.02 ^{B}	0.93 ± 0.02 ^{B}	3.06± 0.02 ^{AB}	3.30 ± 0.05 ^{BC}	85.3 ± 3.5 ^{B}	2.41 ± 0.02 ^{A}
225	**18**	0.90 ± 0.03 ^{D}	0.57 ± 0.03 ^{D}	2.36 ± 0.02 ^{E}	4.15 ± 0.18 ^{A}	65.3 ± 9.6 ^{D}	1.81 ± 0.05 ^{C}
	22	0.99 ± 0.04 ^{CD}	0.66 ± 0.04 ^{CD}	2.36 ± 0.07 ^{E}	3.59 ± 0.23 ^{AB}	69.3 ± 8.7 ^{D}	1.96 ± 0.09 ^{BC}
	26	1.36 ± 0.03 ^{AB}	1.03 ± 0.03 ^{AB}	2.97 ± 0.07 ^{B}	2.87 ± 0.04 ^{DC}	85.3 ± 3.5 ^{B}	2.56 ± 0.05 ^{A}
250	**18**	1.10 ± 0.01 ^{C}	0.77 ± 0.01 ^{C}	3.67 ± 0.07 ^{C}	3.48 ± 0.10 ^{B}	80.0± 6.9 ^{C}	2.13 ± 0.05 ^{B}
	22	1.45 ± 0.02 ^{A}	1.13 ± 0.02 ^{A}	3.12 ± 0.05 ^{AB}	2.76 ± 0.01 ^{D}	88.0 ± 4.6 ^{AB}	2.67 ± 0.01 ^{A}
	26	1.42 ± 0.03 ^{A}	1.08 ± 0.04 ^{A}	3.18 ± 0.05 ^{A}	2.94 ± 0.05 ^{D}	92.0 ± 2.3 ^{A}	2.55 ± 0.05 ^{A}
Pooled Means							
200		1.06 ± 0.01 ^{H}	0.73 ± 0.01 ^{H}	2.71 ± 0.01 ^{H}	3.77 ± 0.01 ^{G}	76.44 ± 0.34 ^{H}	2.08 ± 0.01 ^{H}
225		1.08 ± 0.01 ^{H}	0.76 ± 0.01 ^{H}	2.56 ± 0.01 ^{H}	3.54 ± 0.02 ^{G}	73.33 ± 0.58 ^{H}	2.11 ± 0.01 ^{H}
250		1.33 ± 0.01 ^{G}	0.99 ± 0.01 ^{G}	2.99 ± 0.01 ^{G}	3.06 ± 0.01 ^{H}	86.67 ± 0.36 ^{G}	2.45 ± 0.01 ^{G}
	18	0.98 ± 0.01 ^{Z}	0.65 ± 0.01 ^{Z}	2.50 ± 0.01 ^{Z}	3.89 ± 0.01 ^{X}	72.44 ± 0.50 ^{Y}	1.94 ± 0.01 ^{Z}
	22	1.14 ± 0.01 ^{Y}	0.82 ± 0.01 ^{Y}	2.69 ± 0.01 ^{Y}	3.44 ± 0.02 ^{Y}	76.44 ± 0.49 ^{Y}	2.19 ± 0.01 ^{Y}
	26	1.35 ± 0.01 ^{X}	1.02 ± 0.01 ^{X}	3.07 ± 0.01 ^{X}	3.04 ± 0.01 ^{Z}	87.56 ± 0.23 ^{X}	2.51 ± 0.01 ^{X}

Values represent means of N = 3 replicates / treatment. Means (±SE) in the same row having the same superscript are significantly different (P < 0.01).

1. FCR = feed / gain.

The results of fish performance and body composition for striped mullet ***M. cephalus*** (0.173g initial BW) obtained by El-Dahhar (2000 c) are in general agreement with those obtained for the same fish of nearly similar size 0.2g initial weight (El-Dahhar, 2000 b). However, a diet containing 22% crude protein with 250 kcal ME /100g resulted in greater survival, growth and feed utilization. While, a protein content of 26%, when the energy level was 245 kcal ME /100g, was found to be optimal for striped mullet ***M. cephalus*** in the previous study. Higher availability of protein mixture of El-Dahhar, (2000 c) to the fish, which had a slightly different composition than the previous one (shrimp meal: soybean meal: fishmeal ratio of 1: 1: 1 compared to 1: 2: 1 ratio of the previous study) could be a reason for the difference.

It appears therefore that striped mullet *M. cephalus* required less protein and energy than other fish species studied so far. Further experiments would clarify the relative importance of various energy sources for survival, growth and feed utilization.

A comparison between results of protein and energy maintenance and maximum growth requirements of some fish species is shown in Table 8. Some differences between results were found to be due to different fish species and different experimental conditions e.g. fish size, feeding rate, rearing temperature and others. The smaller fish required more than larger fish due to the higher metabolic rate (Halver, 1988). Alexis and Papaparaskeva-Papoutsoglou, (1986) reported a decrease in the daily protein and energy requirements of ***M. capito*** from 13.5 to 5.3 mg protein /g BW daily and 215.3 to 190.3 cal gross energy /g BW daily as ***M. capito*** increase in BW from 2.5 to 12.7g (Table 8). Also, they recommend a decrease in the optimum dietary protein requirement from 25.5 to 15% as BW increased from 2.5 to 12.7g. El-Dahhar, (1993 and 1994) and El-Dahhar (2000 b) stated that at the same temperature, increasing fish size decreased maintenance and maximum growth requirements for both protein and energy (Table 8). The same author also recommended 52,48, 36, 17.3 and 13.3 mg protein /g BW for 0.2, 0.66, 1.0, 6.3and 9.6 g BW ***O. niloticus***. Hepher ***et al***., (1983) found that specific routine metabolism of red tilapia was 25.4 cal / day at 20.9^{o}C and 36.7 cal / day at 24.3^{o}C. The same author also found that energy maintenance requirement of red tilapia (phenotypically ***Oreochromis niloticus)*** fingerlings 1.0g BW increased from 51 to 73 cal/g BW as water temperature increased from 20.9 to 24.3^{o}C.

Generally, it was concluded that increasing dietary crude protein concentration had a positive effect on weight gain especially in the first stages of growth (Winfree and Stickney 1981; Shiau and Huang, 1989 and 1990; Clark ***et al***., 1990; El-Dahhar, 1994; Twibell and Brown, 1998; El-Dahhar ***et al.*** 1999; El-Dahhar, 2000 a and b). The reason of over estimated values in some of these studies are likely to be due to some factors.

Table 8: *Comparison between results of maintenance and maximum growth requirements of some fish species. Adapted from El-Dahhar (2000 b)*

Fish species	Body weight	Water Tempe -rature	Maintenance Requirements		Maximum Growth Requirements		Dietary Protein Level	Author(s)
			Protein	Energy	Protein	Energy		
	(g)	(°C)	mg/g BW	Cal/g BW	Mg/g BW	cal/g BW	(%)	
Striped mullet	0.19	24.5	3.9	30.2	34.6	345.6	38.0	El-Dahhar, 2000b
Striped mullet	0.2	24.5			26.3	382.7	26.0	El-Dahhar, 2000b
Grey mullet	2.5	23.3			13.5	215.3	25.5	Alexis and Papaparaskeva-Papoutsoglou, 1986.
Grey mullet	12.7	24.0			5.8	190.3	15.0	-Papoutsoglou and Alexis, 1986.
Nile Tilapia	0.2	27.5	10.0	106.9	48.0	513.1	39.6	El-Dahhar, 1993.
Nile Tilapia	0.63	27.0	7.0	76.5	32.0	395	32.0	El-Dahhar *et al.*, 1999
Nile Tilapia	0.76	27.0			30.6		24.0	El-Dahhar *et al.*, 1999
Red Tilapia	1.0	24.3		73.0				Hepher *et al.*, 1983.
Red Tilapia	1.0	20.9		51.0				Hepher *et al.*, 1983.
Nile Tilapia	1.0	27.5	8.6		36.0		30.2	El-Dahhar, 1994.
Hybrid T.	2.9	26.0	4.2		11.9		24.0	Shiau & Huang, 1989.
Nile Tilapia	3.6	27.0			25.8		20.0	El-Dahhar, 2000c
Nile Tilapia	6.3	27.5	4.3		17.5		34.9	El-Dahhar, 1993.
Nile Tilapia	9.6	27.5	3.6		13.3		26.6	El-Dahhar, 1994.
Red Tilapia	11.4	20.0	3.7		42.0		28.2	Zonneveld & Fadholi, 1991.
Catfish	8.5	27.3	1.3	15.1	8.8	99.8	25.0	Gatlin III *et al.*, 1986.
Catfish	9.6	27.3	1.3	15.1	8.8	99.8	35.0	Gatlin III *et al.*, 1986.

Data are based on recalculation of original data.

De-Silva and Anderson, (1995) summarized these factors to be:

1. Energy values of the diets used, are not generally determined but are assumed to be similar to other animals,
2. Inadequate information on amino acids composition of the diets, and
3. Variation in the digestibility of the dietary protein sources

REFERENCES

Albertini-Berhaut, J., 1974. Biology des stages juvéniles de Téléostéens Mugilidae *Mugil auratus* Risso 1810, *Mugil capito* Cuvier 1829 et *Mugil saliens* Risso 1810. II. Modifications du régime alimentaire en relation avec la taille. Aquaculture, 4: 13-27.

Alexis, M. N. and Papaparaskeva-Papoutsoglou, E., 1986. Aminotransferase activity in the liver and muscle of *Mugil capito* fed diets containing different levels of protein and carbohydrate. Comp. Biochem. Physiol. Vol. 83B, No. 1, 245-249.

Baragi, V. and Lovell, R. T. 1986. Digestive enzyme activities in striped bass from first feeding through larva development. Transactions of the American Fisheries Society. 115:478-484.

Benetti D. D. and E. B. Fagundes Netto, 1991. Preliminary results on growth of mullets

(*Mugil liza* and *M. curema*) fed artificial diets. World Aquaculture. 22(4) 1-3.

Blom, J. H. and Dabrowski, K. 2000. Vitamin C requirements of the Angelfish *Pterophylum scalare.* Journal of the World Aquaculture Society. 31(1) 115-118.

Botting, C.C. 1991. Extrusion technology in aquaculture feed processing. Proceedings of the aquaculture feed processing and nutrition workshop. Thailand and Indonesia September 19-25, 1991. Edited by Akiyama, D. M. and R. K. H. Tan. 129-137.

Brusle, J. 1981. Aquaculture of Grey Mullets. Cambridge University Press, IBP 26: 185-212.

Chávez de Martínez, M. C. 1990. Vitamin C requirement of the Mexican native cichlid *Cichlasoma uropthalmus* (Günther). Aquaculture 86: 409-416.

Chávez de Martínez, M. C. and Richards, R. H. 1991. Histopathology of vitamin C deficiency in a cichlid, *Cichlasoma uropthalmus* (Günther). Journal of Fish Diseases 14: 507-519.

Clark J. H. Watanabe, W. O.; Erns, D. H. Wicklund, R. I. and Olla, B. L. 1990. Effect of feeding rate on growth and feed conversion of Florida red tilapia reared in floating marine cages. Turnal of the world Aquaculture Society.21: 16-24.

Dabrowski, K. and Gogowski, J. 1977a. Studies on the proteolytic enzymes of invertebrates constituting fish food. Hydrobiologia 52:171-174.

Dabrowski, K. and Gogowski, J. 1977b. The role of exogenic proteolytic enzymes in digestion processes in fish. Hydrobiologia 54:129-134.

Dabrowski, K. and Gogowski, J. 1977c. A study of the application of proteolytic enzymes to fish food. Aquaculture 12:349-360.

Dabrowski, K. Grudniewski, C. and Dabrowski, K., 1979. Artificial diets for common carp: effect of the addition of enzyme extracts. Progressive Fish-Culturist 41:196-200.

Dabrowski, K. Lee, K. AND Rinchard, J., 2003. The smallest vertebrate, teleost fish, can utilize synthetic dipeptides-based diets. J. Nutr. 133: 4225-4229, 2003.

De la Higuera, M., Murillo, A., Verela, G. and Zamora, S. 1977. The influence of high dietary fat levels on protein utilization by the trout (*Salmo gairdeneri*). Comp. Biochem. Physiol, 56A, 37-41.

Da Silva, S. S. and T. A. Anderson, 1995. Fish Nutrition in Aquaculture. 1st edition. London, Glasgow, Weinheim, New York, Tokyo, Melbourne, Madras.

Degani, D., Ben-Zvi, Y. and Levannon, D. (1989). The effect of different protein levels and temperature on feed utilization, growth and body composition of *Clarias gariepinus* (Burchell 1822). Aquaculture, 76: 293-301.

Devaneson, K. and Chaco, V. 1943. On the possibility of culture of certain marine mullets in fresh water tanks. Proc. Nat. Inst. Sc. India. 9(2): 249-250.

El-Dahhar, A. A. 1993. Maintenance and maximum requirements for fry and fingerlings of Nile tilapia (Oreochromis niloticus) fed at varying feeding regimes. Alexandria Journal of Agriculture Research. 38 (3) : 179 -197.

El-Dahhar, A. A. 1994. Protein requirements of fry and fingerlings of Nile tilapia (Oreochromis niloticus) fed at varying protein level in Egypt. Journal of Agriculture Science, Mansoura University. 19 (1): 117 - 128.

El-Dahhar, A. A, 2000 a. Dietary Vitamin C Requirements of Striped Mullet *Mugil cephalus* Wild Larvae. Conference of Social and Agriculture development in Sinai. 38-46.

El-Dahhar, A. A, 2000 b. Protein and energy requirements of striped mullet *Mugil cephalus* larvae. J. Aric. Sci. Mansoura Univ., 25 (8): 4923 – 4937.

El-Dahhar, A. A. 2000 c. Effect of dietary energy and protein levels on survival, growth and feed utilization of striped mullet *Mugil cephalus* larvae. J. Aric. Sci. Mansoura Univ., 25 (8): 4987 – 5000.

El-Dahhar, A. A and R. T. Lovell, 1995. Effect of protein to energy ratio in purified diets on growth performance and feed utilization of Mossambique tilapia (Oreochromis mossambicus). Aquaculture Research. 26, 451 - 457.

EL-Dahhar, A. A., 1999. Effect of Heat Treated Feed and Exogenous Zymogen® on Survival and Growth of Grey Mullet Larvae *Liza ramada* (Risso) in Egypt. Aquaculture Research. 30, 165-173.

El-Dahhar, A. A.; Moustafa, Y. T.; Salama, M. A. and Zahran, S. 1999. Effect of salinity and salinity acclimation on survival and growth of larval mullet. Alexandria Journal of Agriculture Research.

El-Sayad Rania, M. 2001. Studies on the effect of some vitamins on growth and survival of fish larvae. M. Sc. thesis submitted to Alexandria University. Alexandria, Egypt.

El-Sayed, A. F. and S. I. Teshima. 1992. Protein and energy requirements of Nile tilapia *Oreochromis nilotica* fry. Aquaculture 103:55-63.

Garling, D. L. and R. P. Wilson 1976. The optimum protein to energy ratio for channel catfish *Ictalurus punctatus*. Journal of Nutrition, 106: 1368.

Gatlin III, D. M.; Poe, W. E. and Wilson R. P. 1986. Protein and energy requirements of fingerling channel catfish for maintenance and maximum growth. Journal of Nutrition. 116: 2121-2131.

Govoni, J. J.; Boehller, G. W. and Watanabe, Y., 1986. The physiology of digestion in fish larvae. Environmental Biology of Fishes, 16:59-77.

Govoni, J.J., 1980. Morphological, histological and functional aspects of alimentary canal and associated organ development in larval *Leiostomus xanthurus*. Rev. Can. Biol. 39, 69-80.

Gosline, W. and Brock, V., 1965. Hand book of Hawaiian Fishes, University of Hawaiian Press, Honolulu. cited progp. Fish culture. 36(2) 327p.

Guinea, J. and Fermandez, F. 1991. The effect of SDA, temperature and daily rhythm on the energy metabolism of the mullet *Mugil saliens*. Aquaculture, 97: 353-364.

Halver, J. E., editor. 1988. Fish nutrition, 2nd edition. Academic press, London, England.

Hardie, L. J., Fletcher, T. C. and Secombes, C. J. 1991. The effect of dietary vitamin C for intensification of the Atlantic salmon (*Salmo salar* L.). Aquaculture 95:201-214.

Helland, S. J. and Gisdale-Helland, B. 1998. Growth, feed utilization and body composition of juvenile Atlantic Halibut (*Hippoglossus hippoglossus*) fed diet differing in the ratio between the macro nutrients. Aquaculture 166:49-56.

Hepher, B.; Liao, I. C.; Cheng, S. H. and Hsieh, C. S., 1983.Food utilization by red tilapia – effect of diet composition, feeding level and temperature on utilization efficiencies for maintenance and growth. Aquaculture. 32: 255-275.

Hepher, B.; Liao, I. C.; Cheng, S. H. and Hsieh, C. S., 1983.Food utilization by red tilapia – effect of diet composition, feeding level and temperature on utilization efficiencies for maintenance and growth. Aquaculture. 32: 255-275.

Hilton, J. W., Cho, C. Y. and Slinger S. J. 1978. Effect of graded level of supplemental ascorbic acid in practical diets fed to rainbow trout (*Salmo gairdneri*). Journal of the Fisheries Research Board of Canada 35:431-436.

Jaffa, M. 1989. Vitamin C can curb those stress associated losses. Fish Farmer 12:18-19.

Jantrarotai, W., Stasit, P., Jantrarotai, P., Viputhanumas, T. and Srabua, P. 1998. Protein and energy levels for maximum growth, diet utilization, yield of edible flish and protein sparing of hybrid *Clarias* catfish (*Clarias macrocephalus* X *Clarias gariepinus*). Journal of the World Aquaculture Society, 29(3): 281-289.

Jürss, K. 1981. Influence of temperature and ratio of lipid to protein in diets on aminotransferase activity in the liver and white muscel of rainbow trout (*Salmo gairdneri* Richardson). Comp. Biochem. Physiol., 68B, 527-533.

Kolkovsky, S.; Tandler, A.; Kissil, G. W. and Gertler, A. 1990. The use of exogenous digestive enzymes to improve microdiets for gilthead sea bream (*Sparus aurata*) larvae rearing. Presented at the International Symposium on Development and Aquaculture of Marine Larvae held in Bergen, Norway, 12-15 August 1990.

Landolt, M. L. 1989. The relationship between diet and immune response of fish. Aquaculture, 79: 193-206.

Lauff, M. and Hofer, R. 1984. Proteolytic enzymes in fish development and the importance of dietary enzymes. Aquaculture 37:335-346.

Lee, C. S., and Menu, B., 1981. Effect of salinity on egg development and hatching in grey mullet, *Mugil cephalus* L. Journal of Fish biology. 19: 179-188.

Lee, D. J. and Putnam, G. B., 1973. Response of rainbow trout to varying protein/energy ratio in a test diet. Journal of Nutrition, 103:916-922.

Li, M. H., Wise D. J. and Robinson, E. H. 1998. Effect of vitamin C on weight gain, tissue ascorbate concentration, stress response, and disease resistance of Channel catfish *Ictalurus punctatus*. Journal of the World Aquaculture Society 29(1): 1-8.

Lim, C. and Lovell, R. T. 1978. Pathology of the vitamin C deficiency syndrome in Channel catfish *Ictalurus punctatus*. Journal of Nutrition 108:1137-1146.

Machiels, M. A. M. and Henken, A. M. 1985. Growth rate, feed utilization and energy metabolism of the African catfish, *Clarias gariepinus* (Burchell, 1822) as affected by dietary protein and energy content. Aquaculture, 44: 271-278.

Masumoto, T., Hosokawa, H. and Shimeno, S. 1991. Ascorbic acid’s role in aquaculture nutrition. Proceedings of the aquaculture feed processing and nutrition Workshop, Thailand and Indonesia, September 19-25, 1991: 42-48.

Murashige, R.; Bass, P.; Wallace, L.; Molnar, A.; Eastham, B.; Sato, V.; Tamaru, C. and Lee, C. S., 1991. The effect of salinity on survival and growth of striped mullet (*Mugil cephalus*) larvae in the hatchery. Aquaculture. 96: 249-254.

Nash,C, E. and Shehadeh, Z. H. (Editors), 1980. Review of breeding and propagation techniques for grey mullet, *Mugil cephalus* L. pp. 1-10.

NRC (National Research Council), 1993. Nutrition Requirements of Fish. National Academy Press Washington, D. C., USA.

O'Connell, C.P., 1981. Development 0f organ system in the northern anchovy *Engraulis mordax*, and other teleosts. Am. Zool. 21, 429-446.

Papaparaskeva-Papoutsoglou. E. and Alexis, M. N. 1986. Protein requirements of young grey mullet, *Mugil capito.* Aquaculture, 52: 105-115.

Person Le Ruyet, J.; J. C. Alexandre ; L. Thébaud and C. Mugnier. 1993. Marine fish larvae feeding: Formulated diet or live prey? Journal of the World Aquaculture Society 24(2): 211-224.

Pillai, T. G., 1975. Possibility for aquaculture in Tunisia. Symp. On Aquaculture in Africa. FAO/CL, FA-75 / SE 216.

Reis,L. M., Reutebuch, E. M. and Lovell, R. T. 1989. Protein-to-energy ratios in production diets and growth, feed coversion and body composition of channel catfish *Ictalurus punctatus*. Aquaculture, 77: 21-27.

Sandnes, K. 1991. Vitamin C in fish nutrition a review. Fiskeridirektoratets Skrifter Serie Ernoering 4:3-32.

Sato, M., Kondo, T., Yoshimaka, R., and Ikeda, S. 1982. Effect of dietary ascorbic acid levels on collagen formation in rainbow trout, Bull. Jpn. Soc. Scient. Fish. 48:553-556.

Sauberlich, H. E. 1984. Ascorbic acid. Pages 260-280 in Olson, R. E., Broquist, H. P., Chichester, C. O. Drby, W. J., Kolbye A. C. Stalvey, R. M., editors. Nutrition reviews, present knowledge in nutrition. The Nutrition Foundation, Inc., Washington, D. C., USA.

Sealey, W. M. and Gatlin D. M., III. 1999. Dietary vitamin C requirement of hybrid striped bass *Morone chrysops* (female) × *M. saxatilis* (male). J. World Aquaculture Society. 30 (3) 297-301.

Segner, H., Storch, V., Reinecke, M., Kloas, W., Hanke, W., 1994. The development of functional digestive and metabolic organs in turbot, *Scophthalmus maximus. Mar. Biol. 119, 471-468.*

Serrano, J. A., Nematipour G. R. and Gatlin III, D. M. 1992. Dietary protein requirement of red drum (*Sciaenops ocellatus*) and relative use of dietary carbohydrate and lipid. Aquaculture 101: 283-291.

Shiau, S. Y. and Jan, F. L. 1992. Dietary ascorbic acid requirement of juvenile tilapia *Oreochromis niloticus* × *O. aureus*. Aquaculture, 133:147-157.

Shiau, S. Y. and Huang, S. L., 1989. Optical dietary protein level for hybrid tilapia (*O. niloticus* x *O. aureus*) reared in seawater. Aquaculture, 81: 119-127.

Shiau, S. and Peng, C. 1993. Protein-sparing effect by carbohydrates in diets for tilapia, *Oreochromis niloticus* ×*O. aureus.* Aquaculture, 117: 327-334.

Sylvester, J. R.; Nash, C. E. and Emberson, C. R., 1975. Salinity and oxygen tolerances of eggs and larvae of Hawaiian striped mullet, *Mugil cephalus* L. Journal of Fish Biology. 7: 621-629.

Tanaka, M. 1969. Studies on the structure and function of the digestive system in teleost larvae: II. Characteristics of the digestive system in larvae at the stage of first feeding. Jpn. J. Ichthyol. 16,41-49, (in Japanese, with English abstract).

Tandler, A. and S. Kolkovsky. 1991. Rates of ingestion and digestibility as limiting factors in the successful use of microdiets in *Sparus aurata* larval rearing. European Aquaculture Society 15:169-171.

Twibell, R. G. and P. B. Brown, 1998. Optimal dietary protein level for hybrid tilapia *Oreochromis niloticus* X *O. aureus* fed all-plant diets. Journal of the World Aquaculture Society 29(1):9-16.

Vallet, F., Berhaut, J., Leray, C., Bonnet, B. and Pic, p., 1970. Preliminary experiments on the artificial feeding of Mugilidae. Helgol. Wiss. Meeresunters., 20: 610-619.

Walford, J. and T. J. Lam. 1993. Development of digestive tract and proteolytic enzyme activity in seabass (*Lates calcarifer*) larvae and juveniles. Aquaculture 109: 187-205.

Walford, J. and T. J. Lam. 1993. Development of digestive tract and proteolytic enzyme activity in seabass (*Lates calcarifer*) larvae and juveniles. Aquaculture 109: 187-205.

Walsh, W. A.; Swanson, C.; Lee, C. –S.; Banno, J. E. and Eda, H., 1989. Oxygen consumption by eggs and larvae of striped mullet, *Mugil cephalus*, in relation to development, salinity and temperature. Journal of Fish Biology. 35: 347-358.

Watanabe, T., 1977. Sparing action of lipids on dietary protein in fish-low protein diet with high calorie content. Technocrat, 10 (8): 34-39.

Watanabe, T., Takeuchi, T. and Ogino, C. H. 1979. Studes on the sparing effect of lipids on dietary protein in rainbow trout (*Salmo gairdneri*). In Finfish Nutrition and Fishfeed Technology (Edited by Halver J. E. and Tiews K.), Vol. I, pp. 113-125. Heeneman, Berlin.

Watanabe, T.; Kitajima, C. and Fujita, S., 1983. Nutritional values of live organisms used in Japan for mass propagation of fish: a review. Aquaculture, 34: 115-143.

Whitehead, P. J. P.; Bauchot, M. L.; Hureau, J. C.; Nielsen, J. and Tortonese, E., editors. 1986. Fishes of the North-Eastern Atlantic and the Mediterranean. (Vol3).

Wilson, R. P. and Poe, W. F. 1973. Impaired collagen formation in the scorbutic channel catfish. Journal of Nutrition 103:1359-1364.

Winfree, R. A. and R. R. Stickney, 1981. Effects of dietary protein and energy on growth, feed conversion efficiency and body composition of *Tilapia aurea*. Journal of Nutrition 111:1001-1012.

Zonneveld, N. and Fodholi, R., 1991. Feed intake and growth of red tilapia at different stoking densities in ponds in Indonesia. Aquaculture, 99: 83-94.

استعراض مرجعي علي أقلمة يرقات أسماك البوري وتقدير احتياجاتها الغذائية

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تم استعراض لدراساتنا علي أقلمة يرقات أسماك العائلة البورية خلال فترة العشر سنوات السابقة في هذه المقالة. فقد تم دراسة تأثير ملوحة الماء ونقطة بداية الأقلمة من الصفر إلي ٣٥ جزء في الألف علي حيوية ونمو أسماك البوري ومنها وجد أن أعلي معدل لنفوق الأسماك سجل مع الملوحات المرتفعة (٢٠ و٣٥ جزء في الألف) وأيضا مع الملوحات المنخفضة (صفر جزء في الألف) . من هذا البحث تم التوصل إلي التوصية بنقل اليرقات البرية من أسماك العائلة البورية إلي المعمل في ملوحة مشابهة للملوحة التي انتقلت منها لمدة ٢٤ ساعة بعدها يتم في اليوم التالي أقلمة الأسماك علي الملوحات الأقل.ويساعد ذلك في توضيح نقص أسماك العائلة البورية في ماء البحر المتوسط المالحة. وعادة تختلف اليرقات اختلافا جوهريا عن آبائها في بساطة القناه الهضمية وهذا يعني نقص شديد في الإفرازات الإنزيمية. وفي بحث آخر، لدراسة تأثير الغذاء المعامل حراريا والإنزيمات الخارجية علي حيوية ونمو أسماك الطوبارة grey mullet (*Liza ramada* (Risso, 1826). ومن ثلاثة تجارب وجد أن الإنزيمات الخارجية من الفرائس الطبيعية لها أهمية كبيرة في نشاط هضم البروتين في اليرقات وكذلك الإنزيمات الإضافية لها نفس التأثير، وهذا التأثير هو هضم الغذاء وتنشيط إفراز الأنزيمات الداخلية بانشقاق الصورة الغير نشطة من الإنزيم. وقد تركزت الدراسة في هذا البحث علي تحسين المنتج والاستخدام الأمثل للمواد الخام. ويمكن للمعاملة الحرارية تعقيم المنتج وإيجاد الفرصة لاستخدام المدى الواسع من المواد الخام وبالتالي تعظيم الاستفادة التغذوية منها. وفي دراسة علي تقدير الاحتياجات من الفيتامينات قد تم تقدير الاحتياجات من مخلوط الفيتامينات والأملاح المعدنية وكذلك فيتامين ج لأسماك البوري striped mullet (*Mugil cephalus*)، من هذا البحث توصلنا إلي أن أعلي معدل نمو وحيوية قد وجد مع المعاملات التي تم استخدام مستوى ٠.٩ و ١.١ % من المخلوط التجاري للفيتامينات والأملاح المعدنية وأن أقل مستوى لفيتامين ج هو ٤٠ ملجم / كجم من العلف. العامل التجاري الهام الذي يحدد إنتاج الأسماك في المزارع المكثفة هو العلائق التي تساعد في تعظيم معدل النمو والتي تأخذ في الاعتبار الاحتياج إلي العناصر والفيتامينات الهامة في المحافظة علي صحة الأسماك. وفي بحث علي تقدير احتياجات اليرقات من البروتين والطاقة استخدم سبع نسب بروتين من ١٤ – ٣٨ % بروتين خام و ٢٤٥ ك كالوري طاقة ميتابولزمية/١٠٠ جم علف أوضحت أن أسماك البوري التي تغذت علي ٢٦% بروتين أعطت أعلي معدل نمو وذلك باستخدام نسبة من الدهن في حدود ٦.٣%. وبالإشارة إلي التفاعل بين الطاقة الميتابولزمية ومستوى البروتين فإن اليرقات أعطت أعلي نمو وأعلي حيوية عندما تغذت علي عليقة تحتوي علي ٢٢% بروتين مع ٢٥٠ ك كالوري من الطاقة الميتابولزمية /١٠٠ جم علف (نسبة الدهن ١٥%) وكان ذلك أعلي من كل النسب البروتينية الأخرى مع كل مستويات الطاقة الميتابولزمية المستخدمة.