

STUDIES ON THE PRODUCTION OF SEA BASS, *Dicentrarchus labrax*, UNDER DIFFERENT REARING AND FEEDING CONDITIONS

Marine fish, sea bass (*Dicentrarchus labrax*), sea bream (*Sparus aurata*) and mullet demonstrated the most important fish species in mariculture programs not only in Egypt, but also in Mediterranean countries. Sea bass and breams, the main species produced, experienced the highest increase in all of key Mediterranean species. Mediterranean offshore mariculture is seen as a way to increase fish production in areas where it would otherwise not be possible, and fulfill the lack between the demands and the production of fish in Egypt.

1- EFFECT OF DIETARY PROTEIN SOURCES ON THE GROWTH PERFORMANCE, FEED UTILIZATION AND PROFITABILITY OF SEA BASS POST LARVAE, *Dicentrarchus labrax*.

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ABSTRACT

Six experimental diets were formulated; a fish meal based control diet (1), and five diets in which 25 % of the fish meal was substituted with either algae, liver of cartilages, clams and liver of beef or shrimp meal (diet 2, 3, 4, 5 and 6, respectively). The diets were fed to sea bass post larvae (*Dicentrarchus labrax*) over a 105-day experimental test period. Growth performance, survival rate and feed utilization parameters . The best in fish fed diet (3) containing 22% liver of cartilages, followed by fish fed diet 5, 6, 4, 1 and 2 respectively. Considerable saving in feed costs was achieved when liver of cartilages was used as a partial dietary substitute inside fishmeal within compound feeds for sea bass post larvae statge, about 22.7%.

INTRODUCTION

European sea bass, *Dicentrarchus labrax*, is a very interesting species for Mediterranean aquaculture, but actually its production is low. Therefore, the development of diets with optimum nutrient content, which allow improving the feeding of this species, will increase their production and economic profit. The protein requirements in the young sea bass are high, 35-50% (Hidalgo *et al.*, 1987; Hidalgo and Alliot, 1988; Hassanen *et al.*, 1997; Zegrari *et al.*, 2000 as well as Peres and Oliva-Teles, 2000), to meet their energy needs (Cowy, 1993) by utilize a significant source of the dietary protein. Fishmeal has always been considered the most satisfactory sources of protein for inclusion in formulation for sea bass feed. Despite this material is limited and their highly costs are continuously affecting in a direct way feeding costs and total production costs in aquaculture. For this reason substitution of fishmeal as a major protein source in aqua feeds remains a major issue.

Therefore, the present work was conducted to study the effect of different dietary protein sources on growth performance, and nutrient utilization and economic profit of sea bass post larvae.

MATERIALS AND METHODS

(I) Experimental studies and techniques

This experiment was conducted during March 26 to July 11, 1998 (105 days nursing period). Sea bass post larvae stage were obtained from El-Tarh Private Marine Hatchery, 20 Km in the East of Alexandria, Behera Governorate, in March 11, 1998. They were transported to Fish Nutrition Lab, NIOF, in aerated polyethylene bags and stocked into 500(l) for each fiberglass tanks to realize adaptation in indoor nursing conditions for about two weeks. Twelve experimental glass aquaria, (100cm length, 30 cm in width and 40 cm in depth) were used in the present experiment. Water volume in each aquarium was adjusted at 100 (l) of sea water, which was filtered, and continuously aerated using central air blower. Water in each aquarium was partially changed once every three days but the fecal material was siphoned out daily morning. Healthy sea bass post larvae (with an average weight of 0.04 g and length 1.40 cm) were stocked in the twelve experimental glass aquaria at a rate of 20 post larvae / aquarium,

Six protein sources (fishmeal, appetizers algae, liver of cartilages, clams, liver of beef and shrimp meal) were used for formulating six experimental diets. The diets were isonitrogenous and isocaloric. Formulation and chemical analysis (%) of the tested diets are presented in table (1). Composition of vitamin and mineral premixes (Pfizer Medicines Company) are presented in Tables (2) and (3), respectively. Fish were fed three times daily (9.00 A.M., 15.00 P.M. and 20.00 P.M.) to satiation, for 6 days in week. The fish were considered satiated when they did not show an interest in feed. The mean consumption (appetite) for each treatment was computed for the experimental period of 105 days.

(II) Water quality:

Dissolved oxygen was determined using Winkler's technique (Huet, 1972) and pH was measured using a portable pH meter (Drion Research Model 210). Ammonia, was determined according to the methods described by Golterman *et al.*, (1978).

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Table (1): Composition and proximate analysis of the tested diet proposed for sea bass, *Dicentrarchus labrax* advanced fry, expressed as % of dry weight.

Ingredients	Diet No.					
	1	2	3	4	5	6
Composition (%)						
Fish meal	40	40	30	30	30	30
Soybean meal	20	15	20	20	20	20
Wheat milling by- products	15	10	10	10	10	10
Cod oil	5	5	3	3	5	5
Starch	15	5	10	5	10	10
Appetizers Algae	-	20	-	-	-	-
Liver of Cartilages	-	-	22	-	-	-
Clams	-	-	-	27	-	-
Liver of Beef	-	-	-	-	20	-
Shrimp meal	-	-	-	-	-	20
Vitamin premix	2	2	2	2	2	2
Mineral premix	3	3	3	3	3	3
Proximate analysis						
Dry matter(DM)	91.8	67.4	72.2	76.2	70.2	74.2
Crude protein(CP)	48.02	47.94	48.12	47.93	48.1	47.91
Ether extract(EE)	11.8	11.87	15.11	13.09	13.09	13.48
Crude fiber(CF)	8.1	17.3	6.3	6	6	7.23
Ash	10.2	10.6	11.6	10.4	10.23	11.48
Nitrogen free extract(NFE)	21.88	12.29	18.87	22.58	22.58	19.9
Calculated gross energy (Kcal GE/100g DM)	474.7	434.7	493.9	489.3	492.3	481.7
P/E ratio	10.11	11.02	9.74	9.79	9.74	9.94

Table (2) : Composition of the vitamins * :

Vitamins	Contents and units
Vitamin A	2,200,000 I.U.
Vitamin D ₃	1,100,000 I.U.
Vitamin E	1,500 I.U.
Vitamin K	800 mg
Vitamin B ₁	1,100 mg
Vitamin B ₂	200 mg
Pantothenic acid	5,000 mg
Nicotinic acid	4,000 mg
Vitamin B ₆	2,000 mg
Vitamin H	15 mg
Vitamin B ₁₂	4 mg
Vitamin C	3,000 mg

*Contained in each 1000 g .

Table (3): Composition of the minerals mixture*

Minerals	Contents and units
Iron	160 mg
Manganese	30mg
Magnesium	334 mg
Copper	21.6 mg
Zinc	21.6 mg
Selenium	25 mg
Cobalt	2.38 mg
Iodine	30 mg
Amino acid concentrate	20 g

*Contained in each 1000 g .

(III) Biochemical analysis:

At the end of experiment, diets and fish samples were frozen kept for chemical analysis. The frozen samples were dried at 70°C and passed through a meat mincer into one composite homogenate per group. Contents of homogenized samples were analyzed for protein (Kjeldhal method), fat (ether extract method, Soxhelt), moisture (oven drying) and ash (burn oven) using procedures of Association of Official Analytical Chemists (AOAC, 1995). Energy content (100 g) was estimated by the following equation:
Energy content=(Protein contentx5.64)+(lipid content x 9.44)+ (carbohydrates content x 4.11).

(IV) Growth performance:

At the end of the experiment, several measurements namely growth and feed utilization parameters were calculated as mentioned in Ballestrazzi *et al.*, (1994) as follows:

1. Daily weight gain = Final body weight (g)- Initial body weight (g)/ Days
2. Condition factor (K) = Weight (g)/ (Total length cm)³ x100
3. Specific growth rate (SGR) % = (Ln W₂ - Ln W₁) x 100/ T
Where: W₂ = Weight at the end of the experiment.
W₁ = Weight at the beginning of the experiment.
T = Time (days).
4. Survival rate = No. fish harvested / No. of fish stocked x 100
5. Feed conversion ratio (FCR) = Feed intake (g)/ Weight gain (g)
6. Protein efficiency ratio (PER) = Weight gain (g)/ Protein intake (g)
7. Protein productive value (PPV) = Protein increment (g) / Protein intake (g) x100

Where protein increment is the protein content in fish carcass at the end—at the start of the experiment.

(V) Economical Evaluation:

The economical evaluation was estimated during the first experiment regarding the feed costs in nursing period. The following proposed equations were used in such evaluations (Hassanen, 1997):

Costs of 1 Kg (LE) =Σ of each Kg ingredients x % of each ingredients in diets.
Feed costs per Kg fresh fissile (LE) = Costs per Kg diet (LE) x Feed conversion ratio.

(VI) Statistical Analysis:

Statistical Analysis were performed by using a computer software program Graph PAD instate (Version 2.01) copyright © 1990-1993 Steve Whetzel, Park- Davis 930762 A. Graph PAD software.

The differences between means calculated according to Duncan (1955)

RESULTS AND DISCUSSION

1. Water criteria:

Water quality is one of prime importance in evaluating and regulating its suitability for fish rearing (Boyd, 1979). Water temperature, dissolved oxygen, pH, salinity and ammonia (NH₃) were monitored during the investigation period and the results are summarized in Table (4).

Table (4): Water quality criteria in indoor glass aquaria during the first experiment(March,26 - July 11,1998).

Items	Minimum	Maximum	X ¹ ± SE
Temperature (C°)	19	25	22 ± 3.01
Dissolved oxygen(mg/l)	5.25	7.71	6.53 ± 0.87
pH	7.79	8.1	8.00 ± 0.11
Salinity (ppt)	32.18	36.00	32.81 ± 0.57
Ammonia NH ₃ (mg/l)	0.071	0.11	0.087 ± 0.02

The present results showed a minimum water temperature of 19°C in March and a maximum of 25°C in July, 1998, with an average of 22.00 ± 3.01°C (Table 4). These temperature ranges was beneficial to fish growth and agree with the findings of Claireaux and Lagardere (1999). They reported, the highest metabolic rate of European sea bass (*Dicentrarchus labrax*) was at temperature 25°C, but at water temperature lower than 14°C the active metabolic rate was dropped gradually. The average dissolved oxygen content in the experimental aquaria varied between 5.25 and 7.71 mg/l, with an overall average of 6.53± 0.87 mg/l (Table 4). This range was desirable in fish rearing ponds. Thetmeyer *et al.*, (1999) found that, sea bass (*Dicentrarchus labrax*) need a dissolved oxygen concentration of at least 4.0 mg/l or greater to maintain good health and feed conversion. pH is a measure of the relative acidity of the water. The desirable range of pH for sea bass, *D. labrax* between 7.5 and 8.2 (Pillay, 1990). The pH values in the present study indicated that, the water was always on the alkaline side, 7.79 - 8.10 (Table 4). Water salinity in experimental glass aquaria ranged between 32.18 to 36.33 ppt. This level classifies such water as saline water (Table 4). Ammonia (NH₃) is the primary nitrogenous waste produced by fish from protein digestion and is toxic to fish (Masser, 1997). Results in table(4) indicated that, ammonia concentration in water glass aquaria varied between 0.071 and 0.110 mg/l. This range is beneficial for Juvenile sea bass (*Dicentrarchus labrax*) and agrees with the findings of Tudor *et al.*, (1994). They found that, for juvenile sea bass, sub-lethal levels of ammonia is 0.6-2.0 mg/l, and is known to cause gill and tissue damage, poor growth and increased susceptibility to disease.

2. Growth performance and condition factor:

The effect of additive protein sources on final means of total length, body weight and condition factor (K) of sea bass post larvae stage reared in indoor aquaria for 105 days (nursing period) are presented in Table (5). The results revealed that, the final length, weight and condition factor followed the same trend of variation and indicated that the performance of sea bass post larvae was significantly affected by various protein sources, by comparing the growth of sea bass, it is clear that, the feeding of diet (3), containing liver of cartilages, had the longest body length, and heaviest body weight as well as best condition factor, as compared with those of fry fed other experimental diets. However, growth performance was significantly improved by using other experimental diet 4, 5 and 6 (containing clams, liver of beef and shrimp

meals) as compared to the control group fed on diet (1) with only fish and soybean meals as protein sources. However, the slightly higher final means of length and weight of the control group (diet1) was far from significant as compared to the fish group fed on algal diet (2) (Table 5). It is clear from Table (6) that the fry fed on diet (3), containing liver of cartilages, recorded the highest levels of the average total and daily length gain (5.39 cm and 0.51 mm/day, respectively), as well as average total and daily weight gain (2.49 g and 23.71 mg/day), specific growth rate as compared with those fed on other experimental diets.

Table (5): Mean \pm SE of initial and final body length, weight and condition factor (K) of sea bass, *D. labrax*, post larvae stage fed on 5 diets with different protein sources, during March 26 to July 11, 1998 (105 days nursing period in indoor aquaria)

Items	Diet No.					
	1	2	3	4	5	6
1- Stocking data						
Av. Initial length (cm)	1.40 \pm 0.15	1.40 \pm 0.15	1.40 \pm 0.15	1.40 \pm 0.15	1.40 \pm 0.15	1.40 \pm 0.15
Av. Initial weight (g)	0.04 \pm 0.01	0.04 \pm 0.01	0.04 \pm 0.01	0.04 \pm 0.01	0.04 \pm 0.01	0.04 \pm 0.01
Av. Initial condition factor (k)	1.45 \pm 0.10	1.45 \pm 0.10	1.45 \pm 0.10	1.45 \pm 0.10	1.45 \pm 0.10	1.45 \pm 0.10
2- Harvesting data *						
Av. Final length (cm)	5.44 \pm 0.10 ^c	5.52 \pm 0.10 ^c	6.70 \pm 0.12 ^a	6.25 \pm 0.08 ^b	6.46 \pm 0.08 ^{ab}	6.32 \pm 0.12 ^b
Av. Final weight (g)	1.80 \pm 0.52 ^b	1.70 \pm 0.06 ^b	2.53 \pm 0.06 ^a	2.08 \pm 0.06 ^a	2.50 \pm 0.07 ^a	2.48 \pm 0.06 ^a
Av. Final condition factor (k)	2.48 \pm 0.06 ^b	1.04 \pm 0.04 ^c	2.80 \pm 0.13 ^a	0.85 \pm 0.01 ^c	0.94 \pm 0.03 ^c	1.00 \pm 0.05 ^c

* Different superscript in the same row, indicates significant differences (P<0.05).

1 control diet 2 Appetizers Algae

3 Liver of Cartilages 4 Clams

5 Liver of Beef 6 Shrimp meal

Table (6): Performance of sea bass, *D. labrax*, fry fed diets different of protein sources, during nursing period.

Items	Diet No.					
	1	2	3	4	5	6
No. of fish	23	15	27	24	26	25
Total length gain (cm)	4.00	4.10	5.39	4.84	5.06	4.92
Daily length gain (mm/day)	0.38	0.39	0.51	0.46	0.48	0.47
Total weight gain (g)	1.76	1.66	2.49	2.04	2.48	2.44
Daily weight gain (mg/day)	16.76	15.81	23.71	19.43	23.62	23.24
Specific growth rate (SGR%)	3.63	3.57	3.95	3.76	3.94	3.93
Total fish yield gain (g/100 l)	40.48	24.9	66.42	48.96	64.48	61.00

It is worth mentioning that, the substitution of part of the fish meal with liver of cartilages significantly improved not only the growth performance parameters and condition factor of sea bass, *D. labrax*, fry.

These results might be attributed to one or more of the following factors:

(1) The particulate diet (3) contain proteins and other ingredients that may be easy to be digested by fry. Kolkovaski (2001) reported that, for juvenile fish the enzymes activity has been observed to be relatively low compared with adult fish levels. Moreover, particulate diets for larvae must contain

ingredients with low dry matter content; (2) n-3 highly unsaturated fatty acids (n-3 HUFA) levels in cartilages fish were extremely high (more than 1.3g/Kg on dry weight basis) (Mourente and Odriozola, 1990 and Abi-ayad *et al.*, 1997). An increase in dietary n-3 HUFA levels in diets significantly improved larval growth. This may be due to the greater need of larvae for n-3 HUFA in their metabolism and for membrane construction in accordance with their higher growth rate (Izquierdo and Fernandez-Polacios, 1997; Furuita *et al.*, 1999 and 2000). Bruce *et al.*, (1999) demonstrated that, the larval development and growth of European sea bass (*D. labrax*) were improved by increased levels of both amino acids (AA) and n-3 HUFA in their diet. and (3) sea bass larvae fed the 48% crude protein obtained from cartilages fish and herring fish meals had the highest energy protein and lipid retention. The investigators attributed these results to a higher ability of flatfish elongate and de-saturated fatty acids in comparison with other fish (Izquierdo and Fernandez-Palacios, 1997). In addition to the important conclusion that the percentage of carbohydrate in the present sea bass diet not exceed 20%, lipid content 15% and protein content 48%. These levels were lead to the increase of beneficial for post larvae stgsge growth (Perez *etal.*, 1997; Peres and Oliva-Teles, 1999).

3. Survival percentage:

As previously observed in the effect of the additive protein sources on the growth performance of sea bass (*Dicentrarchus labrax*) post larvae, the survival rate showed the same trend of trend. So the fry fed on diet (3), containing liver of cartilages had the highest survival rate, 90% followed by diets 5 , 6 , 4 , 1 and 2; (Table7).

Table (7) : survival percentage of sea bass , *Dicentrarchus labrax* fry fed diets with different protein sources, during nursing period in indoor aquaria.

Diet No.	No. of fish stocked / 100 (l)	No. of fish harvested / 100(l)	Survival rate (%)
1	30	23	76.66
2	30	15	50.00
3	30	27	90.00
4	30	24	80.00
5	30	26	86.66
6	30	25	83.33

Several authors have reported a reduction in survival rates were recorded when larvae of halibut (Holmefjord and Olsen, 1991), turbot (Gatesoupe and Le Milinaire, 1985), striped jack (Izquierdo , 1988), red sea bream (Izquierdo *et al.*, 1989a and 1989 b) and gilthead sea bream (Rodriguez *et al.*, 1993; 1994; Salhi *et al.*, 1994) were fed low n-3 HUFA levels in their diets (0.3-0.95 g/Kg on dry weight basis). The reflection of weak

swimming ability of fish larvae fed diets of low n-3 HUFA was also demonstrated by Watanabe and Kiron (1994). In relation to the obtained corresponding results of survival percentage of group 3 compared with other groups; it could be mentioned that the relation between n-3 HUFA levels in diets and survival percentage was clearly demonstrated and agrees with the findings of many authors.

4. Feed utilization parameters:

The results obtained for total weight gain and diets utilization (feed conversion ratio, protein efficiency ratio (PER) and protein productive value (PPV%) during the first experiment are shown in Table (8). Sea bass (*Dicentrarchus labrax*) fry utilized artificial diet (3) containing 22% liver of cartilages was better than other groups fed on diets contained in addition to fish and soyabean meals the other sources of protein: as well as algal meal (20%). The feed conversion ratio (FCR), protein productive value (PPV %) and protein efficiency ratio (PER) of fry fed on diet (3) were greatly improved by 4.65%, 6.56% and 33.33%, respectively, in relation to the control (diet 1). The lowest results were obtained of fry fed on diet (2) containing 20% algal meal. n-3 highly unsaturated fatty acids (n-3 HUFA) are one nutritional factor which greatly affects fish growth and diet utilization of marine fish larvae (Furuita et al., 2000). This may be due to the greater need of larvae for n-3 HUFA in their metabolism (Izquierdo and Fernandez-Palacios, 1997). Thereby, the liver of cartilages diet improved diet utilization by sea bass fry due to their high level of n-3 HUFA (Mourente and Odriozola, 1990).

Table (8): Feed utilization parameters: [feed conversion ratio (FCR), protein efficiency ratio (PER), protein productive value (PPV %)], of sea bass, *Dicentrarchus labrax* fry fed on diets differ of protein sources, during nursing period in indoor basins.*

Diet No.	Total weight gain (g/m ³)	Total feed consumed (g/m ³)	Feed conversion ratio (FCR)	PROTEIN UTILIZATION	
				PPV (%)	PER
1	40.48 ^a	87.13 ^a	2.15 ^a	0.61 ^a	0.97 ^a
2	24.90 ^b	51.31 ^b	2.06 ^b	0.48 ^b	1.00 ^b
3	66.42 ^c	136.50 ^c	2.06 ^b	0.65 ^a	1.01 ^b
4	48.96 ^a	103.46 ^c	2.11 ^a	0.60 ^a	0.98 ^b
5	64.49 ^c	138.40 ^c	2.15 ^a	0.60 ^a	0.97 ^a
6	61.00 ^c	135.60 ^c	2.22 ^c	0.55 ^{bc}	0.93 ^c

* Different superscript in the same column, indicates significant differences (P < 0.05)

5. Economical evaluation:

The economical evaluations of the six experimental diets are shown in Table (9). It is important to mention that the final cost of the fish produced was determined by both the feed cost per Kg diet and the feed conversion ratio (weight gain/feed consumed). The differences in feed conversion between six diets resulted in a lowest cost/Kg fish produced with the liver of cartilages diet (3), about 3.08 LE/Kg fish, followed by control diet (1), about 3.96 LE/Kg fish produced (Table 9).

In conclusion, the present study indicated that sea bass fry utilized artificial diet (3), containing 22 % liver of cartilages, better than the expensive control diet (1), containing fish and soybean meals, and possessed best performance and economic profit.

Table (9) : Feed cost per kg of sea bass, *D. labrax* , fry fed on the experimental diets for 105 days in indoor aquaria .

	experimental diets					
	1	2	3	4	5	6
Cost per kg diet ,LE	1.84	1.60	1.5	2.92	3.71	3.91
Consumed feed to produce one kg fish kg	2.15	2.06	2.06	2.11	2.15	2.22
Feed cost / kg fresh fish fissile , LE	3.96	3.30	3.09	6.16	7.94	8.68

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دراسات على إنتاج أسماك القاروص تحت نظم تربية و تغذية مختلفة
١- تأثير اختلاف المصادر البروتينية على معايير النمو و الإعاشة و الاستفادة
الغذائية و الجدوى الاقتصادية لزريعة أسماك القاروص
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** المعهد القومى لعلوم البحار و المصايد- الأفوشى- الإسكندرية

أجريت هذه الدراسة بهدف إدخال بعض البدائل البروتينية لمسحوق السمك فى
عليقة زريعة أسماك القاروص المحتوية على مسحوق السمك كمصدر رئيسى للبروتين و
دراسة تأثير ذلك على معايير النمو و الإعاشة و الاستفادة الغذائية و تكلفة إنتاج الكيلوجرام
سمك حيث تم اختبار ستة علائق: الأولى قياسية تحتوى مسحوق السمك و فول الصويا
كمصدر رئيسى للبروتين و تم استبدال ٢٥ % من بروتين العليقة (الكنترول) فى العليقة
الثانية التى تحتوى على ٢٠ % طحالب جافة و الثالثة على كبد الأسماك الغضروفية
كأسماك المحرثات بنسبة ٢٢ % و الرابعة على المحاربات بنسبة ٢٧ % و الخامسة على
مسحوق كبد البقر المجمد بنسبة ٢٠ % و السادسة على مسحوق الجمبرى بنسبة ٢٠ % .
و تتميز هذه البدائل بتوفرها فى السوق المحلى بسعر رخيص . تشير النتائج إلى أن يرقات
القاروص حققت أفضل معايير للنمو و الإعاشة و الاستفادة الغذائية عند تغذيتها على
العليقة رقم (٣) المحتوية على كبد أسماك المحرثات تلاها العلائق أرقام ٢،١،٤،٦،٥ على
التوالى و هذا راجع إلى أن كبد أسماك المحرثات غنى بالأحماض الدهنية الأساسية الغير
مشبعة المحفزة للنمو كما أوضحت نتائج التحليل الاقتصادي أن تكلفة الكيلو جرام من
العليقة رقم (٣) حوالى ١,٥ جنيه بينما كانت فى العليقة الكنترول حوالى ١,٨٤ جنيه . و
هذا بالطبع أدى إلى خفض تكلفة إنتاج الكيلو جرام من السمك بنسبة ٢٢ % .