

## Dietary *Spirulina platensis* and *Chlorella marina* Microalgae as Growth Promoters During Weaning Post Larvae of European Seabass, *Dicentrarchus labrax*

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**ABSTRACT:** The present study was carried out to evaluate the effect of dietary *Spirulina platensis* and *Chlorella marina* supplementation on growth performance, feed efficiency, survival (%), apparent digestibility coefficients (ADCs) and economics analysis of European seabass, *Dicentrarchus labrax*, post larvae during 60 -120 days of weaning period in hatcheries.. A total of 180 seabass post larvae with an average body weight of  $0.20 \pm 0.01$  g were divided in the three experimental treatments (three replicates each). The experiment was conducted for 60 days. Three iso-nitrogenous (53.64% crude protein) and iso-caloric (15.67MJ/kg, DM) weaning diets were formulated. The control diet had no *Spirulina platensis* and *Chlorella marina* added. Diets 2 and 3 each were formulated to contain 5% *S. platensis* and 5% *C. marina*, respectively. The final weight (FW), average daily gain (ADG), specific growth rate (SGR) , survival (S%) and proximate body composition of *D. labrax* post larvae were significantly ( $P \leq 0.05$ ) improved in experimental diets supplied with *Spirulina platensis* and *Chlorella marina* microalgae compared to the control group. The same trend was recorded for best feed conversion ratio (FCR), protein efficiency ratio (PER), protein productive value (PPV), energy retention (ER) and energy utilization (EU). The higher significant apparent digestibility coefficients values of dietary protein and lipid were recorded for fish fed *S. platensis* than other treatments. No statistical difference ( $P \geq 0.05$ ) was observed for the influence of dietary *S. platensis* and *C. marina* microalgae on whole body proximate analysis of *D. labrax* post larvae. The change in incidence cost were highly significant ( $P < 0.05$ ) for fish fed control diet compared with diets containing *S. platensis* and *C. marina* tested algae. The results of present study suggested that European seabass, *D. labrax*, post larvae fed diets containing 50 g/kg of *Spirulina platensis* and *Chlorella marina* for 60 days had promote and enhanced growth performance, diet utilization efficiency and dietary nutrients digestibility coefficients.

**Keywords:** European seabass, *Dicentrarchus labrax*, *Spirulina platensis*, *Chlorella marina*, growth performance, body composition

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## INTRODUCTION

Microalgae have been used to enhance nutritional and health performance of fish species as dietary supplements. Many studies publicized status of algae as “super food”, for example, 1–5% microalgae led to enhanced growth, improved protein and immune system (Turner *et al.*, 2002), lipid metabolism (Güroy *et al.*, 2011), antiviral, antibacterial and also improved gut function (Michiels *et al.*, 2011), get better stress resistance (Sheikhzadeh *et al.*, 2012). They act as a good source

of protein, amino acids, fatty acids, vitamins, minerals and biologically active phytochemicals (Becker, 2004; Pulz and Gross, 2004). In most feeding trials, microalgae have been found to increase growth performance, feed utilization, and production systems (Meireles *et al.*, 2008). *Chlorella*, *Spirulina*, *Nannochloropsis*, *Tetraselmis*, *Isochrysis*, *Chaetoceros*, *Pavlova*, *Phaeodactylum*, *Skeletonema* and *Thalassiosira* are the most frequently provides improves fish growth (Spolaore *et al.*, 2006).

Among the main areas of dietary algae research for finfish is the potential for protein replacement by microalgae species (Palmegiano *et al.*, 2008). *Spirulina* is one of the most commonly microalgae used in fish feed; it is a rich source of protein, essential amino acids, essential fatty acids, vitamins, minerals and antioxidant pigments (Nakagawa and Montgomery, 2007). The effects of *Spirulina* on fish growth, feed intake and nutrient utilization have been investigated for several fish species, including red sea bream (Mustafa *et al.*, 1997), Nile tilapia, (Takeuchi *et al.*, 2002), carp (Nandeeshia *et al.*, 2001) and white sturgeon (Palmegiano *et al.*, 2008).

In this context, El-Sayed (1994) found that the growth conversion efficiency of silver seabream, *Rhabdosargus sarba* fed on *Spirulina* meal at  $\geq 50\%$  was usually not different from, those given control diets with solely fishmeal. While, at 75% of *Spirulina*, the growth of it reduces, but feed conversion efficiency still comparable to the control diet, which sharply significantly reduces at 100% inclusion level. Feeding on *Spirulina* improve disease resistance and improvement in survival rate from 15 to 30%. Fish-fed *Spirulina* (4% DW) to juvenile trout produced immunoglobulin expressed in different tissues such as skin epithelial or blood cells (Sayre *et al.*, 2001).

*Chlorella* microalgae, is a single celled algae. *Chlorella* is widely distributed and easily cultured in outdoor ponds (Huo *et al.*, 2012) and accordingly is a quite promising source of protein. Owing to the high nutritional value, *Chlorella* is a nutrient-dense super food that contains 60% protein, 18 amino acids (including all the essential amino acids), and various vitamins and minerals. *Chlorella* has been used as dietary protein sources for marine and freshwater fish to improve weight gain and carcass quality (Badwy *et al.*, 2008; Xu *et al.*, 2014). Most of minerals and vitamins are found in *Chlorella*, including iron, calcium, potassium, magnesium, phosphorous, pro-vitamin A, vitamins C, B1, B2, B5, B6, B12, E and K, biotin, inositol, folic acid, plus vitamins C, E and K. *Chlorella* have a better activity in inhibiting lipid peroxidation (Bengwayan *et al.*, 2010).

**Therefore**, the aim of the present study was to evaluate the effect of dietary *Spirulina platensis* and *Chlorella marina* supplementation as growth promoter on growth performance, feed efficiency, survival (%), apparent digestibility coefficients (ADCs) and economics analysis of European seabass, *D. labrax*, post larvae during 60 -120 days of weaning period in hatcheries.

## MATERIALS AND METHODS

### Rearing techniques

Post larvae of *D. labrax* were purchased from El-Sharif Marine Fish Hatchery (Wadi Mariut), Alexandria, Egypt. On days-60, a total of 180 seabass post larvae with an average body weight of  $0.20 \pm 0.01$  g. were stocking in 9 glass aquaria (30×40×70 cm with capacity of 70 liters) at stock density of 20 post larvae/aquaria representing three experimental groups (in triplicate each). The larvae were fed experimental micro-encapsulation diets four times daily (7.00, 9.00, 11.00 and 15.00 hr). The experiment was conducted for 60 days.

### Preparation of Experimental Microalgae

At the beginning of the trial, 500 g fresh weight of each microalgae (in triplicate each) were collected for determination of initial proximate nutrients composition (Table 1).

### Experimental diets formulation

Three iso-nitrogenous (53.64% crude protein) and iso-caloric (15.67MJ/kg, DM) weaning diets were formulated. The control diet had no *Spirulina platensis* and *Chlorella marina* added. Diets 2 and 3 each were formulated to contain 5% *S. platensis* and 5% *C. marina*, respectively (Table 2).

All experimental diets were formulated using a software diet formulator program using an industrial twin-screw extruder (Beijing modern Yangong machinery S&T development Co. Ltd, China). The processing conditions were as follow: the ingredients were super fine- milled (sieved with screen diameter >50 µm), mixed well and then incorporated before being included in the diets by extruder system. The moisture of the ingredients was increased to 30-40% by adding hot water in the manual conditioner, increasing its temperature to 70-90°C. Time of conditioning was 200–240 s. All diets products were ground through a 2 mm cutter to diet preparation. Then, the diets were dried to 2.7% moisture using a drier (at 60–70°C, 48 h), coated by fish oil, minerals, vitamin premix and additives, cooled and stored frozen (-18°C) until used, according to Goda *et al.* (2017).

The different experimental diets prepared as micro-encapsulation whose size must be adapted to the size of the larvae mouth, where manual crashing and grading (well calibrated to minimize waste) to four size of the micro-encapsulation diets used in our experiments was 300–500 µm from day 60 to day 75, then 500–800 µm to day 80, then 800 – 1000 µm to day 100, finally used 1200 µm to finally experiments (day 120), Transition from one stage to another takes about 5-10 days according to growth.

### Growth Performance, Feed and Nutrients Utilization Parameters

Total post larvae weight was determined to the nearest 0.1 milligrams (Sartorius CP224S – Sartorius AG, Germany), total length was determined to the nearest millimeter and the fish immediately returned to their aquaria conditions. The feed amounts were adjusted and corrected according to their weight. The performance parameters were conducted according to the following equations (Goda et al., 2017):

**Final body weight (LBW, g/fish):** Live body weight (LBW) in g of an individual fish of each experimental treatment was recorded every 15 days and at the end of the trial.

**Weight gain (WG, g/fish)** =  $W_t - W_0$

Where:  $W_t$ : final weight  $W_0$ : initial weight

**Average daily gain (ADG, g/fish/day)**  $(W_t - W_0)/n$

Where:  $W_t$ : final weight;  $W_0$ : initial weight; n: duration period (days)

**Specific growth rate (SGR, %/ day)** =  $100 \times (\ln W_t - \ln W_0) / \text{days}$

Where:  $W_t$ : final weight;  $W_0$ : initial weight; Ln: natural logarithm

**Survival (S, %)**  $S = (\text{No. of fish at end} / \text{No. of fish at the start}) \times 100$

**Total length gain (TLG, cm)** = [Average final length (cm) – Average initial length (cm)].

**Condition factor (K value)** =  $100 \times (W/L^3)$ , where W= weight(g) and L= length(cm)

**Feed intake (FI, g):** This is the amount of feed given or supplied during the experimental period.

**Feed conversion ratio (FCR)** = dry matter intake (g)/ body weight gain (g).

Where: Body weight gain =  $(W_t - W_0)$ .

**Protein efficiency ratio (PER)** = weight gain (g)/ protein intake (g).

**Protein productive value (PPV, %)** =  $100 \times (P_t - P_0) / \text{protein intake (g)}$ .

Where:  $P_0$ : protein content in fish carcass at the start;  $P_t$ : protein content in fish carcass at the end.

**Energy retention (ER, %)** =  $(E_t - E_0) / \text{Energy intake (Kcal)} \times 100$ .

Where:  $E_t$ : energy content in fish carcass (Kcal) at the end;  $E_0$ : energy content in fish carcass (Kcal) at the start.

**Energy Efficiency Ratio (EER)** = (energy weight gain (Kcal) /energy intake (Kcal))  $\times 100$ .

### Analytical methods

At the beginning of the trial, a random pooled of three samples (75 fish) were collected, anesthetized with t-amyl alcohol and sacrificed for determination of initial whole-body proximate composition. At the experiment termination, six fish were randomly selected from each treatment and anesthetized with t-amyl alcohol, sacrificed, and homogenized in a blender for final whole-body proximate composition. The fish were pooled for each aquarium, oven-dried, ground, and stored at  $-20^\circ\text{C}$  for subsequent analysis. The chemical composition of fish and diet samples were determined according to the procedures of AOAC (2000). Dry matter was determined after drying the samples in an oven ( $105^\circ\text{C}$ ) for 24 h. Ash by incineration at  $550^\circ\text{C}$  for 12 h. Crude protein was determined by micro-Kjeldhal

method, %N × 6.25 (using Kjelttech autoanalyzer, Model 1030, Tecator, Höganäs, Sweden) and crude fat by Soxhlet extraction with diethyl ether (40 - 60°C). Gross energy were calculated using gross calorific values of 5.65, 9.45 and 4.11 kcal g for protein, fat and carbohydrate, respectively according to NRC (1993).

### Digestibility Determination

Before the end of experimental and for ten days later, fish fecal was collected from the each experimental water aquaria's groups by siphoned technique. The fecal were dried properly using in an oven at 105°C overnight. Dietary acid insoluble ash (AIA) was used as a marker, according to (NRC, 1993). The percent nutrients digestibility was determined using the following formula: Digestion coefficients of nutrition

$$= 100 - \frac{100(\text{present marker in feed} * \text{precent nutrition in feces})}{(\text{precent marker in feces} * \text{precent nutrition in feed})}$$

### Economical Evaluation

The economical evaluation of different experimental diets was done including the feed and post larvae cost. The cost of experimental diets was calculated in L.E. according to the local market prices at year 2017 using procedure of El-Dakar *et al.* (2007).

**Table (1). Proximate analysis of experimental microalgae**

| Parameters                           | <i>C. marina</i> | <i>S. platensis</i> |
|--------------------------------------|------------------|---------------------|
| Dry matter (%)                       | 92.80            | 93.58               |
| <b><u>On dry matter base(%):</u></b> |                  |                     |
| Crude protein                        | 34.56            | 62.71               |
| Ether extract                        | 9.57             | 6.70                |
| Total carbohydrate                   | 29.00            | 20.94               |
| Ash                                  | 26.09            | 9.65                |

**Table (2). Composition and proximate analysis of experimental diets**

| Ingredients (g)                      | Experimental diets |                  |                     |
|--------------------------------------|--------------------|------------------|---------------------|
|                                      | Control            | <i>C. marina</i> | <i>S. platensis</i> |
| Fish meal (65% - Yemen) <sup>1</sup> | 480                | 480              | 480                 |
| Meat & bone meal (48%) <sup>2</sup>  | 60                 | 60               | 60                  |
| Soybean meal (48 solv) <sup>3</sup>  | 180                | 180              | 180                 |
| Gluten (corn) <sup>4</sup>           | 70                 | 70               | 70                  |
| Wheat Bran (14 CP) <sup>5</sup>      | 70                 | 20               | 20                  |
| Corn (7.5% CP)                       | 50                 | 50               | 50                  |
| Microalgae                           | 0                  | 50               | 50                  |
| Fish oil <sup>7</sup>                | 70                 | 70               | 70                  |
| Mono-Calcium Phosphate               | 10                 | 10               | 10                  |
| Vitamin premix <sup>8</sup>          | 2                  | 2                | 2                   |
| Trace mineral premix <sup>8</sup>    | 2                  | 2                | 2                   |
| Vitamin C <sup>9</sup>               | 2                  | 2                | 2                   |
| Garlic <sup>11</sup>                 | 2                  | 2                | 2                   |
| Methionine <sup>10</sup>             | 1                  | 1                | 1                   |
| Lysine <sup>10</sup>                 | 1                  | 1                | 1                   |
| <b>Chemical composition (%)</b>      |                    |                  |                     |
| DM                                   | 90.47              | 90.84            | 90.83               |
| <b>On dry matter base (%):</b>       |                    |                  |                     |
| CP                                   | 52.41              | 53.56            | 54.96               |
| Lipid                                | 15.43              | 15.78            | 15.67               |
| Fiber                                | 2.03               | 2.11             | 2.04                |
| Ash                                  | 10.37              | 11.62            | 10.86               |
| GE kCal/kg <sup>12</sup>             | 20.27              | 19.46            | 19.46               |
| DE kCal/kg*                          | 15.35              | 14.78            | 14.78               |

<sup>1</sup>Triple Nine fish protein, Denmark; <sup>2</sup>International Feed, Daniels Street, Long Lake, MN 55356 U.S.A.; <sup>3</sup>Alexandria Company for the manufacture of seed, Egypt; <sup>4</sup>American corn gluten, USA; <sup>5</sup> El Nasr Mill, Egypt; <sup>7</sup>Crude Sardine Oil, INDIA; <sup>8</sup> vitamin and mineral Premix Composition:- Each 3 kg contains : Vit A (6000000 IU.), Vit D (900000 IU), Vit E (10000 mg,) Vit K3 (1000 mg) Vit B1 (1000 mg), Vit B2 (4000 mg), Vit B6 (1000 mg), Vit B12 (10 mg), Choline chloride (25000 mg), Folic acid (1000 mg), Biotin (25mg), Pantothenic acid (8000 mg), nicotinic acid (20000 mg), Magnesium sulphate (20000 mg), Copper sulphate (5000 mg), Iron sulphate (250000 mg), Zinc sulphate (4000 mg), Cobalt sulphate (100mg), manganese (40000 mg), iodine (500 mg), selenium (100 mg), Carrier calcium carbonate up to 3000 mg – produced by AGRE - VET for manufacturing vitamins and feed additives, Egypt; <sup>9</sup>CUXA-VIT C, German; <sup>10</sup>ALPHOS®, Belgium, <sup>11</sup>the Egyptian French factory, Egypt.

<sup>11</sup> Nitrogen free extract % calculated by differences= 100- (CP%- Lipid%- Fibre%- Ash%); <sup>12</sup>GE: gross energy calculated on the basis of 23.6, 39.4 and 17.2 k joule GE g-1 protein, ether extract and total carbohydrates, respectively (NRC, 1993).

### Statistical Analysis

Data were statistically analyzed by ANOVA using SPSS Statistics 17.0 procedure. The assay data were submitted to Bartlett test to verify homoscedasticity. The data showed no variances in homogeneity. Subsequently, the data were submitted to one ways classification variance analysis. Duncan's

multiple range test was used to compare differences between treatment means when significant F values were observed (Duncan, 1955), at ( $P \leq 0.05$ ) level.

## RESULTS

### Growth performance

The average of initial weight (IW, g/fish), final weight (FW, g/fish), weight gain (WG, g/fish), average daily gain (ADG, g/fish/day), specific growth rate (SGR, %/day), survival rate, initial length (IL, mm/fish), final length (FL, mm/fish), length gain (LG, mm/fish), condition factor (K value) of *D. labrax* post larvae fed microalgae (*Chlorella marina* and *Spirulina platensis*) are presented in Table (3).

All treatments nearly have the same initial body weight which ranged between 0.19 – 0.21 g, confirming the appropriate of randomization process. This created a suitable condition to appraise the effect of dietary treatments on the performance of fish during subsequent periods of the all feeding trails (60 days). The obtained result showed significant ( $P < 0.01$ ) differences in FW, WG, ADG, SGR between experimental fish groups. The highest significant ( $P < 0.01$ ) values of FW ADG, SGR and SGR were recorded for fish fed *S. platensis* diet, compared to the lowest significant ( $P < 0.01$ ) values recorded for fish fed control diet. The same trend was observed for the survival (%). the results showed that both tested microalgae induced the IL, FL, LG and condition factor (K value) values (Table 4) compared to the lowest values which recorded for post larvae fed control diet.

### Feed utilization

Table 5 shows the results of feed utilization of *D. labrax* larvae feeding diets containing *S. platensis* and *C. marina*. The highest FCR and PER, PPV and EU values were recorded for post larvae fed the diets containing *S. platensis* and *C. marina*. The results revealed that post larvae fed the diets containing *S. platensis* and *C. marina* had enhanced ER compared to control diet.

### Proximate body composition

The body composition analysis of *D. labrax* e.g. dry matter (DM %), crude protein (CP %), ether extract (EE %) and (ash %) are presented in Table (6). Generally, the body composition analysis was differed in (CP% and EE %) between the initial and final body composition. No significant difference in CP%, EE % DM% and Ash% between all experimental fish groups.

### Apparent Digestibility coefficients (ADCs)

Table (7) shows the changes in apparent digestibility coefficients (ADCs) including experimental dietary protein, lipid and carbohydrate contents. The results showed that *S. platensis* diet recorded the highest significant ADC% values of protein compared with other treatments. The highest significant ( $P < 0.05$ ) values of ADC% values for lipid was recorded in *S. platensis* and control diets, respectively. On the other hand, the highest values of carbohydrate digestibility recorded in *Spirulina platensis* diet, compared with the lowest value of control diet.

**Table (3). Growth performance and survival rate of *D. labrax* fed experimental diets containing *S. platensis* and *C. marina***

| Parameters       | Experimental diets       |                           |                          |
|------------------|--------------------------|---------------------------|--------------------------|
|                  | Control                  | <i>C. marina</i>          | <i>S. platensis</i>      |
| IW (g/fish)      | 0.19±0.01                | 0.21±0.01                 | 0.21±0.01                |
| FW (g/fish)      | 1.46±0.03 <sup>c</sup>   | 1.98±0.12 <sup>ab</sup>   | 2.16±0.11 <sup>a</sup>   |
| WG (g/fish)      | 1.27±0.01 <sup>c</sup>   | 1.77±0.112 <sup>ab</sup>  | 1.95±0.03 <sup>a</sup>   |
| ADG (g/fish/day) | 0.021±0.001 <sup>c</sup> | 0.030±0.018 <sup>ab</sup> | 0.033±0.006 <sup>a</sup> |
| SGR (%/day)      | 3.37±0.03 <sup>c</sup>   | 3.72±0.07 <sup>ab</sup>   | 3.90±0.06 <sup>a</sup>   |
| Survival (%)     | 83.33±1.67 <sup>b</sup>  | 93.33±3.33 <sup>a</sup>   | 95.00±2.89 <sup>a</sup>  |

\*Values (mean ± S.E. of three replications) in the same rows having different superscripts letter are significantly (P<0.01).

\*\*Initial weight (IW) - Final weight (FW) - Weight gain (WG) - Average daily gain (ADG) - Specific growth rate (SGR).

**Table (4). Length performance of *D. labrax* fed experimental diets containing *S. platensis* and *C. marina***

| Parameters                 | Experimental diets      |                          |                         |
|----------------------------|-------------------------|--------------------------|-------------------------|
|                            | Control                 | <i>C. marina</i>         | <i>S. platensis</i>     |
| Initial length (mm/fish)   | 18.33±0.67              | 19.00±0.58               | 18.33±0.67              |
| Final length (mm/fish)     | 41.67±0.88 <sup>c</sup> | 49.33±0.33 <sup>a</sup>  | 50.67±1.86 <sup>a</sup> |
| Length Gain (mm/fish)      | 23.33±1.45 <sup>c</sup> | 30.33±0.67 <sup>ab</sup> | 32.33±2.19 <sup>a</sup> |
| Condition factor (K value) | 3.51±0.08 <sup>c</sup>  | 4.02±0.24 <sup>a</sup>   | 4.27±0.19 <sup>a</sup>  |

\*Values (mean ± S.E. of three replications) in the same rows having different superscripts letter are significantly (P<0.01)

**Table (5). Feed utilization of *D. labrax* fed experimental diets containing *S. platensis* and *C. marina***

| Parameters | Experimental diets     |                          |                         |
|------------|------------------------|--------------------------|-------------------------|
|            | Control                | <i>C. marina</i>         | <i>S. platensis</i>     |
| FCR (g)    | 3.00±0.10 <sup>c</sup> | 1.97±0.07 <sup>ab</sup>  | 1.90±0.01 <sup>a</sup>  |
| PER (g)    | 0.65±0.02 <sup>c</sup> | 0.98±0.03 <sup>ab</sup>  | 1.01±0.01 <sup>a</sup>  |
| PPV (%)    | 9.02±0.17 <sup>c</sup> | 14.49±0.90 <sup>ab</sup> | 15.41±0.58 <sup>a</sup> |
| ER (%)     | 9.43±0.12 <sup>c</sup> | 15.34±0.95 <sup>a</sup>  | 16.00±0.61 <sup>a</sup> |
| EU (%)     | 7.48±0.23 <sup>c</sup> | 11.85±0.42 <sup>ab</sup> | 12.34±0.08 <sup>a</sup> |

\*Values (mean ± S.E. of three replications) in the same rows having different superscripts letter are significantly (P<0.01).

\*\*Feed conversion ratio (FCR) - Protein efficiency ratio (PER) - Protein productive value (PPV %) - Energy retention (ER) - Energy utilization (EU)

**Table (6). Proximate body composition of *D. labrax* larvae feeding diets containing *S. platensis* and *C. marina***

| Parameters        | Experimental diets |                  |                     |             |
|-------------------|--------------------|------------------|---------------------|-------------|
|                   | Control            | <i>C. marina</i> | <i>S. platensis</i> | Initial     |
| Dry matter (%)    | 24.14±0.80         | 25.32±0.78       | 25.32±0.78          | 23.49±0.003 |
| Crude protein (%) | 62.15±0.29         | 60.38±0.66       | 61.60±0.13          | 59.08±0.164 |
| Ether extract (%) | 18.17±0.49         | 17.94±0.08       | 17.33±0.21          | 16.26±0.154 |
| Ash (%)           | 16.86±0.64         | 16.05±0.23       | 16.46±0.63          | 19.87±0.268 |

\*Values (mean ± S.E. of three replications) in the same rows having different superscripts letter are significantly (P<0.05)

**Table (7). Apparent digestibility coefficients of *D. labrax* larvae fed diets containing *S. platensis* and *C. marina***

| Apparent Digestibility Coefficients (ADCs) | Experimental diets      |                         |                         |
|--|-------------------------|-------------------------|-------------------------|
|  | Control                 | <i>C. marina</i>        | <i>S. platensis</i>     |
| ADC% protein                               | 88.17±0.63 <sup>c</sup> | 93.50±0.02 <sup>b</sup> | 95.76±0.02 <sup>a</sup> |
| ADC% Lipid                                 | 93.77±0.81 <sup>a</sup> | 90.25±0.26 <sup>b</sup> | 92.38±0.14 <sup>a</sup> |
| ADC% Carbohydrate                          | 60.32±1.04 <sup>c</sup> | 73.01±3.46 <sup>b</sup> | 81.80±0.58 <sup>a</sup> |

\*Values (mean ± S.E. of three replications) in the same rows having different superscripts letter are significantly (P<0.05).

**Table (8). Cost-profit analysis of *D. labrax* fry different experimental diets containing *S. platensis* and *C. marina***

| Parameters                       | Experimental diets         |                             |                             |
|----------------------------------|----------------------------|-----------------------------|-----------------------------|
|                                  | Control                    | <i>C. marina</i>            | <i>S. platensis</i>         |
| Feed price <sup>1</sup> (LE/kg)  | 14.50                      | 14.25                       | 14.25                       |
| Incidence cost <sup>2</sup> (LE) | 1255.14±1.88 <sup>a</sup>  | 1249.37±1.52 <sup>c</sup>   | 1252.73±1.18 <sup>b</sup>   |
| Change in incidence cost (%)     | 100.00±0.15 <sup>a</sup>   | 99.55±0.12 <sup>c</sup>     | 99.82±0.09 <sup>bc</sup>    |
| Value of fish <sup>3</sup> (LE)  | 2500.00±50.00 <sup>b</sup> | 3266.67±116.67 <sup>a</sup> | 3325.00±101.04 <sup>a</sup> |
| Profit index <sup>4</sup> (LE)   | 1.99±0.04 <sup>c</sup>     | 2.61±0.10 <sup>ab</sup>     | 2.65±0.08 <sup>a</sup>      |
| Change of profit index (%)       | 100.00±2.15 <sup>c</sup>   | 131.27±4.82 <sup>ab</sup>   | 133.26±4.17 <sup>a</sup>    |

\*Values (mean ± S.E. of three replications) in the same rows having different superscripts letter are significantly (P<0.05).

1. Costs were as common commercial feeds in international and local markets (Prices in LE).
2. Incidence cost = feed cost to produce 1000 fry of fish + fish price (1200 LE/Fry as 60 day after hatch).
3. Value of fish = price of 1000 fry (3000 L.E. > 1.5 g & 3500 L.E. <1.5 g) \* Survival.
4. Profit index = value of fish /cost of feed consumed, (1000 fish).

### Cost benefits analysis

The enterprise budgets for production of *D. labrax* using different experimental treatments are presented in Table (8). The results indicated that, the incidence cost and change in incidence cost were highly significant ( $P < 0.05$ ) for fish fed control diet compared with the other diets containing different algae. The values of fish, profit index and change of profit index were highly significantly ( $P > 0.05$ ) for fish fed difference *S. platensis* and *C. marina* compared with fish fed control diet.

## DISCUSSION

In the present study, *D. labrax* post larvae feeding micro-encapsulation weaning diets supplementation of 5% microalgae either *Spirulina platensis* or *Chlorella marina* improved weight gain, specific growth rate, feed conversion ratio and protein efficiency ratio of juvenile *D. labrax*. Growth performance and survival rate (S %) were improved for larvae fed the diets supplied with 5% *Spirulina platensis* or 5% *Chlorella marina*, respectively compared to the control larvae group. The highest significant ( $P < 0.01$ ) values of FW ADG, SGR and SGR were recorded for fish fed *S. platensis* diet. The best FCR and the highest PER, PPV, ER and EU were recorded for fish fed the diets containing *S. platensis* and *C. marina*. Also, there was no significant ( $p > 0.05$ ) difference in P%, EE % DM% and Ash% between all analyzed fish treatments. The highest significantly ( $P < 0.05$ ) difference values for ADC% of protein and lipid digestibility recorded in *S. platensis* diet. It's clear that *S. platensis* and *C. marina* diets caused highly significant increase in ADC% of protein digestibility compared to control diet. The results indicated that, the incidence cost and the change in incidence cost were highly significant ( $P < 0.05$ ) for fish fed compared with control diet.

Abdel-Tawwab and Ahmad (2009) reported that fish fed a diet enriched by *Spirulina* (5.0 g/ kg) showed significantly better growth and feed utilization as compared with fish fed the control diet. The dietary supplementation of live *Spirulina* could enhance fish growth and immunity similar to other organisms such as brewer's yeast (*Saccharomyces cerevisiae*), which had been reported to enhance the growth and immunity of Nile tilapia (Lara-Flores *et al.*, 2003; Abdel-Tawwab *et al.*, 2008). These results agree with those reported by Hayashi *et al.* (1998), Hirahashi *et al.* (2002), they reported that feeding *Spirulina* to fish improved the survival and growth rates. Watanabe *et al.* (1990) mentioned that feed supplemented with *Spirulina* powder improved the feed conversion ratio and growth rates for striped jack, *Pseudocaranx dentex*.

Nakagawa *et al.* (1984 & 1983); Nematipour *et al.* (1988) reported that dietary supplementation of 1-5% *Chlorella* improved growth performance, feed utilization, disease resistance and stress response for ayu fish . Also, dietary

supplementation of 5% *Chlorella* meal increased weight gain and feed efficiency ratio in nibbler (Nakazoe *et al.*, 1986).

In the present study, dietary supplementation of 5% *S. platensis* and *C. marina* increased survival in *D. labrax* post larvae. Several studies have been reported to increase survival levels in feed 1-5% *Chlorella* (Nakagawa *et al.*, 1983; Nematipour *et al.*, 1987, 1988, 1990) and red sea bream fed 3% *Spirulina* (Nakagawa *et al.*, 2000). However, the optimum dietary supplementation levels for the positive effects on growth performance may be different among fish species, and the positive effects of 0.5% supplementation might be due to efficacy of extract fraction of *Chlorella*.

The positive effects of *S. platensis* and *C. marina* on growth and feed utilization of *Dicentrarchus labrax* larvae may be related to their valuable chemical contents such as protein, minerals, vitamins, fibre, feeding attractants, antioxidants and unknown growth promoters which have a high nutritional values (Mustafa and Nakagawa 1995; Nakagawa, 1997). Therefore, this study suggests that the *S. platensis* and *C. marina* microalgae had positive effect on apparent digestibility coefficients of dietary nutrients.

## CONCLUSION

The results of the present study has demonstrated that supplementation of 5% *Spirulina platensis* or *Chlorella marina* individually in weaning diet of European sea bass, *Dicentrarchus labrax* larvae act as growth promoters in early weaning larval in marine hatchery management conditions, since they led to improve growth performance, feed utilization efficiency, survival, elevated deposition of protein and less of lipid in body composition, improve apparent digestibility coefficients of dietary nutrients and obtained highest profit index (LE), values for *D. labrax* post larvae nursed in marine hatcheries.

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## الملخص العربي

### تأثير اضافة طحالب السبيرولينا بلانتسيس والكلوريل مارينا علي معدلات النمو خلال مرحلة ما بعد الفطام ليرقات اسماك القاروص الاوروي

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أجريت هذه الدراسة لتقييم تأثير إضافة طحالب سبيرولينا بلانتسيس و الكلوريل مارينا على العلف الماكول وأداء النمو وكفاءة الاستفادة من الغذاء ومعدل البقاء والتحليل الاقتصادي لليرقات (اسماك القاروص) في علائق الاسماك في المفرخات خلال ٦٠ يوم، تم تحضير ثلاثة علائق تحتوي علي (٥٣.٥ ± ١.١% بروتين خام) و دهون (15.67 ±) MJ / kg 0.2 علائق فطام جهزت احداها ككنترول وتم اضافة ٥% من طحلب سبيرولينا بلانتسيس و ٥% طحلب الكلوريل مارينا. تم تقدير معدلات النمو والاعاشة والتحليل الكيماوي للجسم ليرقات اسماك القاروص تم اختبارها للمقارنة مع علائق الكنترول والعلائق المعاملة بالطحالب الدقيقة. افضل معدل تحويل للغذاء ومعدل استفادة من البروتين والطاقة المحتجزة والطاقة المهضومة سجلت للأسماك التي تغذت علي علائق تحتوي الطحالب الدقيقة. وسجلت اعلي معدلات نمو والزياده اليومية في الوزن و معدل الهضم الكلي للبروتين للعلائق التي تغذت علي طحلب اسبيرولينا بالمقارنة بالمعاملات الاخرى. لا يوجد اي اختلاف معنوي بالتحليل الكيماوي للجسم للبروتين والدهون والرماد بين المعاملات المختلفة. تشير هذه النتائج إلى أن إضافة ٥% من المجفف سبيرولينا بلانتسيس و/او الكلوريل مارينا يحسن من معدلات النمو والاستفادة الغذائية ودعلات البقاء وأعلى مؤشر ربح بالجنية من يرقات القاروص الأوروي ، بالإضافة إلى الانخفاض في التكلفة الكلية لانتاج يرقات اسماك القاروص في المفرخات للعلائق الغذائية التي تحتوي على طحالب مختلفة والتي تم اختبارها.

