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Adaptation, growth and bio-economic evaluation of wild-caught salema (*Sarpa Salpa* Linnaeus, 1758) juveniles in culture conditions

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Present study was conducted to provide information on the adaptation capability of salema to culture conditions and evaluate growth performance and bio-economic benefits as a potential candidate for marine aquaculture and alternative to fishing. Wild fish caught using casting nets were fed with trash fish initially until adapted to tank conditions. Thereafter fish were adapted to pellets by gradual replacement of trash feed. Dry feed adapted fish were then fed experimental diets containing four different protein levels (25, 30, 35, and 40%). Overall, salema fed lower protein diets showed better performance and bio-economic results. Broken-line analyses indicated that the optimum protein for best performance were 30.5% under the conditions applied in this study. Finally, salema might be acclimatized to culture conditions and artificial diets, however, further studies are encouraged under different water temperatures with long-term feeding and different stocking densities, artificial spawning and fry production before a conclusion on the feasibility of salema culture can be made.

[**Key words**: Alternative fish, bio-economic evaluation, growth performance, protein, Salema, *Sarpa salpa*]

Introduction

The Mediterranean marine aquaculture industry is in a fast growth period and has doubled its total production from 140.000 tons in year 2000 to about 276.000 tons with annual value of around 1.783.000.000 USD in year 2014, according to the statistics[1]. With this increasing trend, the aquaculture industry seems to be capable to supply and meet an important amount of high quality food demand of the increasing world population that is expected to reach about 8.5 billion in year 2030 and 9.7 billion in 2050[2]. However the two main aquaculture species in the Mediterranean are seabream and seabass, which market price decreased due to the high production of 199.000 tons in Greece and Turkey[1]. Alternative fish species that can be adapted to culture conditions may improve the product diversity and extend market opportunities.

The salema (*Sarpa salpa* Linnaeus, 1758) belonging to the Sparidae family, is a herbivorous species, mainly feeding on aquatic plants and found abundantly on seagrass beds such as *Posedonia sp*. or *Cymodocea sp*. near the shore on sandy or rocky sea bottom[3, 4]. It can be found in various depth ranging from 0 to 70 m in the eastern part of the Atlantic (from the North Sea to Cape of Good Hope, the Canaries and Cape Verde Islands, in the Mediterranean and the Black seas), in the western part of the Indian Ocean (from Mozambique to Cape of Good Hope)[5, 6]. Interestingly, it is reported that in the Mediterranean, salema aggregates around floating cage farms, feeding on uneaten pellets[7], sharing the feed of seabass and seabream. Similar report was published by Neofitou[8], who indicated that pellets in great quantities were found in the stomach of salema (*S. salpa*), white trevally (*Pseudocaranx dentex*) and striped mullet (*Mugil sephalus*) captured around aquaculture cage farms from the Mediterranean. The presence of salema around cage farms gave the idea that it might be potentially acclimatized to farm conditions and to artificial diets. As a demersal marine fish, salema is widely distributed in different areas in the world and the geographic distribution, reproductive biology, age-growth variation in the wild population or the ecological characteristics of this species have been well reported in earlier studies[3, 9, 10, 11, 12, 13]. However, to our knowledge so far, there is no published report available on the growth performance or feed utilization of salema under culture conditions. Therefore, this is the first record for the assessment of adaptation capabilities and suitability of salema to culture conditions with reference to growth performance, feed utilization and bio-economic considerations in recirculating aquaculture systems.

Materials and Methods

Salema juveniles were captured from the wild population on the southern coast of Canakkale Strait (formerly the Dardanelles), Dardanos-Canakkale, Turkey (40°04’30.18”N - 26°21’28.09”E; 40°04’31.07”N - 26°21’25.70”E; 40°04’42.30”N - 26°21’34.58”E; 40°04’41.33”N - 26°21’37.48”E) from April to May in 2014, using casting net and transferred to the marine aquaculture research and development facilities of Faculty of Marine Science and Technology at Canakkale Onsekiz Mart University (Dardanos-Canakkale, Turkey). Fish were distributed in circular polyethylene tanks with aerated seawater and fed with trash fish for a period of two month until the fish adapted to the tank environment. Initially fish preferred to remain close to the tank bottom, and by the end of the tank acclimatization period, fish gradually came to the surface when feed was given. The behavior of heading towards the surface was accepted as a sign of adaptation to the tank environment. Then, trash fish was gradually replaced with pellet feeds. Initially, trash fish was given in the morning time and pellet feed in the evening time. This feeding regime was changed vice versa after a one month feeding, where the pellet diet was given in the morning time and trash fish in the evening, which then in a month later was gradually replaced with pellet feeds only. From the start of the adaptation period to artificial pellet feeding, fish fully adapted to artificial pellet feeds after a period of three month. In a total of five month period, including the first two month of feeding on trash fish, fish became well adapted to both tank environment and artificial pellet diets. Then, experimental fish were mass weighed in buckets with water (10 fish per bucket, initial weight of 17.17±0.08 g) and distributed into 12 circular polyethylene tanks of 100 L (10 fish per tank, 3 replicates per treatment, total fish of 120) in a recirculation seawater system equipped with aeration and biological filtration units. Experimental fish in tanks were weighed at initial and the end of the trial (days 0 and 60). More frequent samplings were not preferred to avoid handling stress as the fish were captured from nature and adapted to culture conditions. Photoperiod regime was constant of 12 h light/12 h dark period. Water parameters such as temperature, salinity, pH, dissolved oxygen were measured daily using a YSI 600 XL MPS multi-probe water analyzer, while unionized ammonia concentrations (NH3-N mg/L seawater) were determined every three days by the Nessler method using a HANNA C200 portable spectrophotometer (HANNA Instruments, Co., Italy) as described by Yigit et al[14], and recorded as 11.43 ± 1.20°C, 28.24 ± 3.99 ppm salinity, 8.17 ± 0.22 pH, 9.26 ± 0.41 mg L^{-1} DO and 0.35 ± 0.12 mg L^{-1} NH_3-N.

Feeding experiment was initiated in December 2014 and lasted for 60 days until February 2015. Totally four fish meal based iso-caloric diets with increasing protein levels from 25, 30, 35, and 40% were formulated and produced using commercial ingredients as shown in Table 1. Prior to pellet production, all the ingredients were mixed with a laboratory mixer, than water was added to the mixture in order to gain a suitable pulp. The well mixed soft pulp was made into a 2 mm pellet size by using a meat grinder and dried at 40 °C in a drying chamber. Experimental diets were placed in a freezer and stored at -20 °C until use.

Fish were hand fed ad-libitum twice a day throughout the feeding trial of 60 days. From the experience and observations during the adaptation period, it was decided that feeding was continuing for 15-20 minutes. This was also followed in the feeding experiment and feed was withheld after 20 minutes of feeding with special attention to avoid overfeeding. The time when fish refused feeding was accepted as a point of satiety. Feed consumption was monitored to be certain of the even distribution of the feed offered by all fish in the experimental tanks, and feed intake was recorded daily by subtracting the feed distributed from the initial weight of feed.

Table 1-Feed ingredients (g/100g) of experimental diets

	Experimental diet / Protein level			
Ingredients (g/100g)	D1/25	D2/30	D3/35	D4/40
Fish meal[1]	41.0	49	56.5	64.7
Corn Starch	5.0	5.0	5.0	5.0
Dextrin	40.5	33.5	26.5	19
Fish oil	9.0	8.0	7.5	6.8
Vit.-mineral mix[2]	4.0	4.0	4.0	4.0
Cholin chloride	0.5	0.5	0.5	0.5
Total	100	100	100	100
Nutritional composition (% dry matter, except for moisture)				
Moisture	8.65	8.44	8.27	8.29
Crude protein	24.88	30.04	35.16	39.66
Crude lipid	12.82	12.65	12.83	12.90
Crude ash	6.54	7.19	8.61	9.38
Crude fiber	3.0	3.0	3.0	3.0
NFE[3]	44.11	38.68	32.13	26.77
GE (kJ/g diet)[4]	18.24	18.45	18.58	18.74
P:E (mg/kJ)[5]	13.64	16.29	18.92	21.17

[1]Brown fish meal, Black Sea Anchovy
[2]Vit.A 65.000 IU, Vit.D3 45.000 IU, Vit.E 25 IU; Vit.K3 5 mg, Vit.B1 12.5 mg, Vit.B2 12.5 mg, Vit.B6 15 mg, Vit.B12 0.025 mg and ascorbic acid 120 mg; Ca 100 mg, P 50 mg, K 30 mg, Na 20 mg, Mg 10 mg, Fe 22 mg, Zn 3 mg, Mn 3 mg, Cu 1.8 mg, Co 0.15 mg, I 0.12 mg, Se 0.05 mg, DL-calcium pantothenate 40 mg, niacin 50 mg, folic acid 2.5 mg, biotin 0.08 mg and inositol 75 mg.
[3]Nitrogen free extracts = dry matter- (crude lipid + crude ash + crude protein)
[4]Gross energy estimated based on 23.6 kJ/g for protein, 39.5 kJ/g for lipid and 17 kJ/g for NFE.
[5]Protein-to-energy ratio in mg/kJ

Results were expressed as mean ± standard deviation (SD) and group mean differences were compared using one-way ANOVA. A significant level of $P < 0.05$ was employed at all cases. For the determination of the optimum dietary protein matching to the maximum specific growth rate, third order polynomial regression between dietary protein and specific growth rate was applied[15].

Results and Discussion

Despite some losses (13.5%) during the acclimatization of salema to the culture conditions, survival rate during the feeding trial was recorded as 100% in all experimental groups showing that all fish were well acclimated to experimental conditions and pellets. During the acclimatization period, it was noted that wild-caught salema was not very resistant to handling or transportation stress. The mortality of 13.5% at the beginning occurred mainly after fish weighing or measuring. When the fish were not handled or weighed, it was seen that the mortality stopped. Due, in the present study, experimental fish were mass weighed using a 10 liter plastic bucket half-filled with seawater.

Average initial weight of 17.17±0.08 g attained a percent weight increase of 7.16±2.48, 13.57±1.71, 10.70±1.62, and 6.20±2.43 % at the end of the 60 days feeding period for the groups fed dietary protein levels of 25, 30, 35, and 40%, respectively. The best growth performance was recorded in the experimental group fed with diets containing 30-35% protein levels. The SGRs, RGRs, and FCRs improved significantly ($p<0.05$) when dietary protein level increased from 25 to 35%, then significantly declined ($p<0.05$) when protein levels in the diets gradually increased to 40% level. Growth and feed utilization was better in the 30% dietary protein group compared to the 35% protein diet, however the difference between these two groups was not significant ($p>0.05$) (Table 2).

The relationship between dietary protein level and the SGR have been used for the estimation of the optimum protein requirements for salema juveniles. From the polynomial regression analyses[15] in Fig.2, it can be assumed that the optimum protein requirement for salema juveniles is about 30.5% of the diet.

The best FCRs were again recorded for the fish in the experimental groups with 30, 35, and 40% dietary protein levels, with no significant differences ($p<0.05$) among these groups (Table 2). Protein efficiency improved with the decrease of dietary protein level, however the difference between the PERs among experimental groups of 25, 30, 35, and 40% dietary protein was not statistically significant ($p>0.05$) (Table 2).

The bio-economic analyses given in Table 3 is also in agreement with the growth performance and feed utilization results in terms of best profit obtained with the 30 % dietary protein level, which was followed by the 35% protein group. However, the feed expenses as percent of profit were similar among the experimental groups of 30, 35, and 40% protein levels ($p>0.05$).

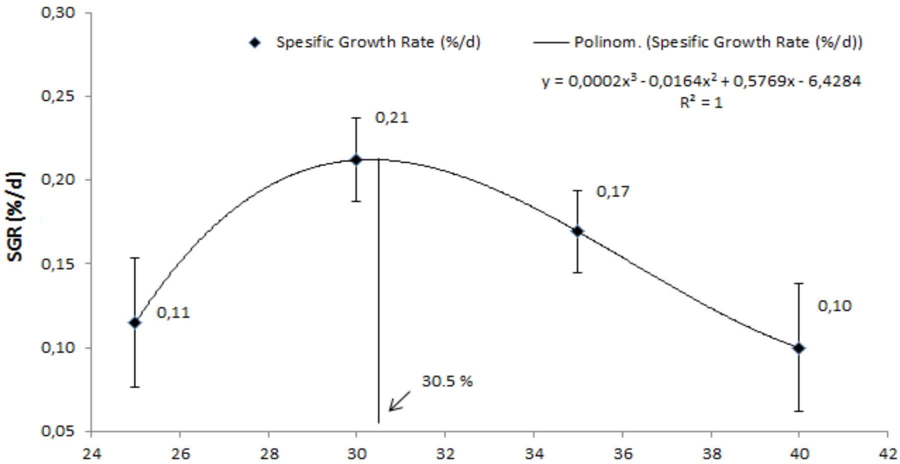

Fig.1-Optimum dietary protein requirement of salema (*Sarpa salpa*) juvenile by polynomial regression between dietary protein levels and specific growth rates

Table 2-Growth performance and feed utilization efficiency of salema (*Sarpa salpa*) juveniles fed different dietary protein levels for 60 days (means ± SD for triplicate groups; values in the same column with the same superscript are not significantly different (p>0.05))

Diets	RGR	SGR	FCR	PER
D1/25	7.16±2.48^{ab}	0.11±0.04^{ab}	1.63±0.08^{b}	2.47±0.12^{b}
D2/30	13.57±1.71^{c}	0.21±0.03^{c}	1.37±0.04^{a}	2.43±0.08^{b}
D3/35	10.70±1.62^{bc}	0.17±0.02^{bc}	1.47±0.13^{a}	1.94±0.17^{a}
D4/40	6.20±2.43^{a}	0.10±0.04^{a}	1.59±0.17^{b}	1.60±0.19^{a}

SGR, specific growth rate (% day^{-1}) = [(ln final weight (g) –ln initial weight (g))/days] x 100
RGR, relative growth rate (%) = [(final wet weight (g)–initial wet weight (g))/initial weight (g)]x100
FCR, feed conversion rate = feed intake (g) / weight gain (g)
PER, protein efficiency rate = wet weight gain (g) / protein intake (g)

Table 3-Bio-economic analyses of salema (*Sarpa salpa*) juveniles fed diets with different levels of protein for 60 days (means ± SD for triplicate groups; values in the same line with the same superscript are not significantly different (p>0.05))

	Experimental diet / Protein level (%)			
	D1/25	D2/30	D3/35	D4/40
Feed supply (kg/fish)	0.020±0.01^{ab}	0.032±0.00^{c}	0.027±0.00^{bc}	0.016±0.01^{a}
Mean weight gain (kg)	0.012±0.00^{ab}	0.023±0.00^{c}	0.018±0.00^{bc}	0.011±0.00^{a}
Feeding cost ($/kg)	0.023±0.01^{a}	0.038±0.00^{b}	0.032±0.00^{ab}	0.025±0.01^{a}
Gross income ($/fish)	0.047±0.02^{ab}	0.089±0.01^{c}	0.070±0.01^{bc}	0.040±0.02^{a}
Σ final biomass cost ($)	0.700±0.02^{b}	0.740±0.01^{c}	0.722±0.01^{bc}	0.693±0.02^{a}
Profit ($/kg)	0.677±0.01^{ab}	0.703±0.01^{c}	0.689±0.01^{bc}	0.669±0.01^{a}

Price of feed varied from 1.15 to 1.60 $/kg from 25 to 50% protein diet; fish sales price was estimated as 3.80 $/kg based on Istanbul actual market price for sparids; other costs than feed expenses are ignored and assumed to be same for all experimental groups.
Feed supply (kg/fish); MWG, mean weight gain (kg)
Feeding cost ($/kg) = feed supply (kg/fish) x feed cost ($/kg)
Gross income ($/fish) = mean weight gain (kg) x price of fish ($/kg)
Σ final biomass cost ($) = final fish weight (kg) x price of fish ($/kg)
Profit ($/kg) = (total final biomass cost - total initial biomass cost) - feeding cost

After introducing wild-caught salema into the culture tanks, fish did not show easy or rapid adaptation to rearing conditions. They were very sensitive to handling procedures such as netting or weighing, and demonstrated a shied swimming behavior in the tank environment, remaining on the tank bottom, that could be due to their native habit since salema is reported as a benthopelagic oceanodromous species, distributed in littoral waters near rocks with *Posidonia oceanica* seagrass beds and sandy or muddy grounds from shallow to 70 m deep water layers[10, 16, 17, 18]. Hence, fish handling was not practical initially, however about 2 month later, salema juveniles showed schooling behavior indicating a state of adaptation to confinement, and fish started swimming to water surface when feed was offered. It was seen that salema became resistant to culture conditions and artificial diets when adequate handling procedures are applied during an adaptation period for 5 month in total, when they showed similar feeding and schooling behavior similar to the other sparids such as gilthead seabream (*Sparus aurata*[19]), axillary seabream (*Pagellus acarne*[20]) or two-banded seabream (*Diplodus vulgaris*[21, 22]). Wild populations of salema are reported to have a mixed diet preference, which is age-related, i.e. where larvae are planktivorous, while young or the adults are herbivorous[23, 24, 25]. There is no published data available on feed requirements or growth performance of salema under culture conditions. However, based on their herbivorous feeding nature, it can be presumed that their dietary protein requirement might be lower than commercially produced carnivorous sparid such as gilthead seabream. The excessive supplement of dietary animal proteins may result in increased nitrogen excretion. Hence, adding dietary animal protein or lipids at an optimum level might support the aquaculture industry in an economic manner and environmental perspective[26]. In the present study, optimum dietary protein requirement of salema juveniles by polynomial regression between dietary protein levels and specific growth rates was found as 30.5% under the conditions applied in the present experimentation.

Our findings showed that a diet containing 30% crude protein gave the maximum growth performance of juvenile salema. In contrast to our findings, dietary protein requirement levels for carnivorous species widely cultured in fish farms, such as gilthead sea bream[27], rainbow trout[28], or European sea bass[29] are reported between 45 and 55%. The result for the dietary protein requirement of salema (*S. salpa*) (30.5%) in the present study is higher than that of an earlier report for white sea bream (*Diplodus sargus*) (27 % protein[30]), but lower than two-banded seabream (*D. vulgaris*) (35-36 % protein[21, 26]), sharpsnout sea bream (*D. puntazzo*) (43-47 % protein[31, 32]), gilthead seabream (*S. aurata*) (50–54 % protein[33]), dentex (*Dentex dentex*) (50% protein[34]), and red progy (*Pagrus pagrus*) (50 % protein[35]). Wilson[36] reported that the optimal protein level in fish diet might be affected by the amino acid composition of the test proteins. Hence, the optimal protein levels even for the same fish species are also subject to differ among various studies. One of the reasons for the lower protein requirement of salema compared to other sparids or other marine species could be due to its herbivorous feeding nature, whereas the other fishes reported to have higher protein requirements in earlier studies are either omnivorous or carnivorous species.

The best FCRs and SGRs in the present study were found for the fish fed the 30 and 35 % protein diets compared to the high protein groups. Similar results to our findings for FCRs (1.37-1.84) in the present study were reported for two banded seabream (*D. vulgaris*) (1.50-1.80[20], 1.36-2.96[21], 1.67-1.92[37]), zebra seabream (1.69-3.33[38]), and gilthead seabream (0.91-3.06[39], 1.14-3.73[40], 1.22-1.74[18], 1.24-1.48[41], 1.37-1.53[42]). In contrast, lower FCRs (1.1-1.2) were reported for gilthead seabream by Bischoff et al[43] in recirculating aquaculture conditions, while higher feed conversion of 2.51 was reported for a new aquaculture species, the axillary seabream (*P. acarne*) in cage environment[19].

The SGR (0.21 % day^{-1}) of salema fed the best performing protein diet (30%) in the present study was similar to the SGR reported for axillary seabream (0.23 % day^{-1}) introduced as a new species for aquaculture[19], but lower than those reported for gilthead seabream (0.32-1.04 %/day[39]). Other slow growing marine fish species, such as white seabream (*D. sargus*) and zebra seabream (*Diplodus cervinus*) were also reported to demonstrate low SGRa (0.89, 0.8 %/day) by Sá et al[29] and Coutinho[38]. In contrast, Ozório et al[25] and Coutinho et al[31] recorded higher SGRs for two-banded seabream, *D. vulgaris* (1.22 %/day) and sharpsnout seabream, *Diplodus puntazzo* (1.54 %/day), respectively, also considered as new candidate species for the marine aquaculture.

A significant decrease in PERs was observed in the present study when dietary protein levels increased. Our finding in terms of decreasing PERs with increasing dietary protein levels agreed with earlier reports in European eel (*Anguilla anguilla*)[44] and in yellow snapper (*Lutjanus argentiventris*)[45], and in two-banded seabream *D. vulgaris*[20].

The discrepancies of results between different studies could be attributed to the rearing conditions such as water temperature, dissolved oxygen, salinity, or fish stocking rates or feeding methods[18, 40], but also to fish species and size of fish[34].

Furthermore, salema juveniles used in this study were wild-caught species and it is likely that their adaptation capability to tank environments were weak, which possibly affected the growth performance and a strong reason for the slow growth rate recorded in the present study. Feeding on pelleted dry diets was conducted for a period of 60 days. Long-term feeding trials are encouraged to collect more information on growth performance of salema of different size groups.

Additionally, the size of the circular shaped experimental tanks was relatively small (100 L) and light green in color. In a recent study, Kesbic et al[37] investigated feed utilization and growth performance of juvenile seabass (*Dicentrarchus labrax*) cultured in tanks with different colors, and reported that both the growth rate and feed efficiency of fish were affected by the color of the tank. The authors reported that the selection of appropriate tank color is important in terms of better growth achievements and economic benefits in aquaculture facilities. Similarly, Howell[46], Chatain and Ounais-Guschemann[47], Rotllant et al[48], Duray et al[49], Strand et al[50], and Imanpoor and Abdollahi[51] also reported that the background color of the tanks used in aquaculture affected the rearing success in turbot (*Scophthalmus maximus*), gilthead seabream (*S. aurata*), red porgy (*P. pagrus*), grouper (*Epinephelus suillus*), Eurasian perch (*Perca fluviatilis*), and caspian kutum (*Rutilus frisii*), respectively.

It is reported that different fish species require diverse environmental ambience throughout their life stage, and that the color of the culture environment may affect fish welfare for a less stressful condition, and since the fish is held in closed containments in aquaculture facilities, which is totally different than their natural environment, feeding activity, growth and fish welfare may be negatively affected under stressful conditions[52, 53]. In the present study, salema was sensitive to handling procedures and showed a shied swimming behavior in the tank environment, resting close to tank bottom, which might be due to their natural habitat in the nature. Matić-Skoko et al[10], Francour[15], Guidetti[16], and Pallaoro et al[17] reported that salema is a benthopelagic oceanodromous species, which population is distributed in littoral waters in rocky sea ground with *P. oceanica* seagrass beds and sandy or muddy grounds. This information gives an explanation for the natural habitat of salema and the light green color of the tanks used in the present study might have negatively affected fish welfare and led to stressful culture conditions. Hence it might be advisable to investigate the color of the tanks best suitable for salema culture.

The bio-economic evaluation of the results showed that the decline or increase of feeding costs were related to the amount of feed consumption, and the protein levels of the test diets. In general, our results in terms of gross income (GI) values were lower compared to the rates given for gilthead seabream and two banded seabream diets in previous studies by Yigit et al[18] and Bulut et al[20], respectively. Findings related to the profit and feed costs as percentage of profit values from the present study were also lower than those reported for gilthead seabream or two banded seabream diets by Yigit et al[18] and Bulut et al[20], respectively. The differences between these studies could be attributed to different fish species, culture conditions, feed quality, type and amount of fish meal sources used in the diets. Overall, the economic analyses results conducted in the present study supports growth parameters found with the highest GI and profit for the diets containing 30 and 35 % proteins.

Conclusion

Based on the findings in the present study, salema demonstrated a none-easy adaptation progress to culture conditions, while its adaptation to artificial diets was much easier. The results from the feeding trial in the present study show that the use of practical diets containing 30-35 % protein provides efficient and cost effective production of salema juveniles compared to the higher protein diets. However, further studies are encouraged on growth performance and nutritional requirements of different size groups of salema, as well as artificial breeding and larvae culture, or health management of salema before a full-developed aquaculture of this fish species could be initiated, since the un-controlled or non-regulated capture based aquaculture will only increase fishing pressure on natural ocean resources due to the efforts of providing juveniles to supply fish farming.

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