Promoting the culture of gilthead sea bream (*Sparus auratus* L.) in low saline inland water:

A novel way to farm saltwater fish in freshwater

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A group of researchers from the Bengis Centre for Desert Aquaculture, Ben-Gurion University of Negev, Sede Boker Campus in Israel has developed a novel technique of culturing saltwater fish in freshwater environment by adding salt in the fish diets. Although developed in Israel, this novel method could also be adopted in the Southeast Asian region to promote the aquaculture of high-value marine fishes in the region's large freshwater bodies paving the way for abating the economic and development disparities in the region's aquaculture industry. The results of the work conducted by the researchers are laid out in this article.

Sea bream (0.4±0.14 g) were reared in three water salinities 3.0, 5.5 and 7.0 ppt by mixing brackish water (2.5 ppt salinity) taken from a local brackish geothermal subsurface aquifer with Dead Sea salt. The 140 days experiment was conducted in 60-liter (l) rearing tanks in a re-circulating system. Three diets were used in the experiment: Diet 1 "Denis premium 1" (54% protein and 14.5% fat; manufactured by RMC Ltd, Israel), which served as the control diet; Diet 2 and Diet 3 which were prepared by adding 0.5% and 1.5% Red Sea salt, respectively to the control feed, fine grounded and re-pelleted. Fish were fed ad libitum five times daily. Fish reared in 3.0 ppt salinity and fed the diet containing 1.5% salt grew significantly ($p<0.05$) better than those fed the control diet. In 5.5 ppt and 7.0 ppt salinities, fish fed the control or the salt rich diets did not differ significantly in growth among the groups regardless of the diets given. Adding 0.5% sea salt to the diet promotes better survival of the fish reared in 3.0 ppt salinity (42.1%) and 7.0 ppt salinity (50.7%). The specific growth rates (SGR) of fish reared in 5.5 ppt and 7.0 ppt salinities and fed the control diets was lower than those fed salt rich diets, although statistically the difference was not significant. In contrast, fish reared in 3.0 ppt salinity and fed the diet containing 1.5% salt had significantly higher SGR than those given the control diet. The present results indicated that the effect of salt-enriched diet on sea bream is more pronounced when reared in lower salinity. Fish culture in low water salinity opens up promising opportunities for the production of sea bream and other euryhaline fish species in arid regions holding brackish water.

Gilthead Sea Bream

Gilthead sea bream (*Sparus auratus* L.) is a euryhaline teleost capable of living in environments of different salinities ranging from 2 ppt to 60 ppt (Laiz-Carrion et al. 2005). *S. auratus* is found in the natural habitat from the Mediterranean and Black Sea to the eastern Atlantic Ocean from Senegal to the United Kingdom (Kissil et al. 2000). It usually inhabits the shallow lagoons along the coasts, but migrates into deeper waters to spawn after late autumn. In commercial rearing units, the early larval stages after yolk absorption (3-4 days) are fed with rotifer (e.g. *Brachionus plicatilis*) followed by *Artemia* after 12-15 days post hatching and this is continued up to 32-35 days post hatching.

The sea bream larvae should be fed with commercial lipids and fatty acids enrichments along with rotifer and Artemia, mainly for normal growth, development and survival. The larvae could then be weaned at 5-10 mg size with 50-60% high protein formulated diet.

Acclimation into Low Saline Waters

Adaptation of euryhaline fish into different environmental salinities induces changes/activation of its iron transport mechanism. This is usually accompanied with changes in oxygen consumption, suggesting variations in the energy demands for osmoregulation (Sangiao-Alvarellos et al. 2003). Sea bream rearing in low saline waters has been successfully tried in many research experiments giving a number of possibilities for culturing the fish in low saline inland waters. This would create an important expansion of aqua-business ventures which can be adapted in many regions. Cultivation practices of sea bream in the Mediterranean region (annual production ~ 150,000 mt) vary due to the region's environmental conditions.

However, since inland low saline water is deficient in its mineral (salt) content, adding the required salt into the diet of the fish could produce better results in terms of growth and survival (Harpaz et al.2005). Sea bream could also be explored as potential candidate species for such culture system using marine, brackish or low saline inland waters. Fish adapted to low saline water exhibits a passive outward flux of ions such as Na+ and Cl- to the water via the gills, faeces, and renal system, which must be compensated by the active uptake of ions (e.g., Na+, Cl-, K+, and Ca2+) from the water and/or from the diet. The diet of fish therefore, constitutes an important source of salts that can satisfy the osmoregulatory requirements particularly when fish are kept in low saline water, thus sparing energy used for osmoregulation and reserving more energy for growth. It has been shown that the salinity of the rearing water has some influence on feed intake in rainbow trout. In high saline water (up to 28 ppt), feed intake usually increases but the growth rate decreases affecting feed conversion ratio. Yet, in euryhaline species, salinity affects growth in various ways and maximum growth is not always obtained under isosmotic conditions.

Salt Addition in Diets

Many researchers have addressed the issue of using dietary salt to help alleviate the problems associated with transferring salmonids to seawater or saltwater growing conditions. The results have shown a marked advantage in the use of dietary salt resulting in better survival. Even the transfer of tilapia, a freshwater (non-anadromous) species to saltwater (15-20 ppt) conditions was easier for the pre-acclimatized fish by adding salt to their diet (Fontainhas-Fernandes et al. 2001). Addition of salt to the diet of fish has several advantages as it increases appetite and also acts as a humectant by reducing water activity. Although the effect on the growth of fish may not be very clear, some studies have attained positive results. Some researchers may have considered this experiment advantageous but others reported that the growth in young coho salmon was hampered even with the addition of only 1.5% salt resulting in a 7% weight gain reduction (Fontainhas-Fernandes et al. 2000; Nandeesha et al. 2000; Eroldogan 2003).

It should be noted that in the analysis of the mineral composition of various fish feeds available in the market, the content of salt in these feeds could be 1.5% on the average but could also be as high as 6.0% especially in the case of feeds used for the early stage of salmon fingerlings. A large percentage of the dietary salt in commercial feeds for carnivorous fishes originates from the fish meal component of the diet. This is one factor therefore, that should be taken into account when replacing fish meal with various plant-derived meals which are not rich in salt. Nonetheless, it should also be considered that better utilization of the food is dependent on the digestion and absorption capabilities of the fish.

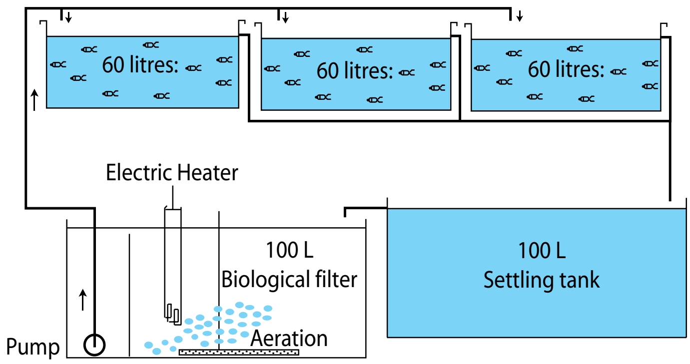

Fig 1: The experimental setup (showing one of three units)

Research Methods and Findings

The nine rectangular rearing tanks (60-l capacity each) used for the experiment were grouped into three separate re-circulating systems with each system consisting of three rearing tanks connected to a separate water-cleaning unit so that all three replicates share the same water parameters through the water circulation unit. The water-cleaning unit (100-l tank), which is filled with volcanic gravel and strongly aerated acts as a biological filter (**Fig. 1**). Filtered water enters each rearing tank at the rate of 3.2 l/min while 15 l of water were replaced daily in each of the three systems.

The salinities of the three systems were prepared by using local brackish water of 2.5 ppt salinity pumped from a brackish geothermal aquifer resting deeply subsurface, into which Dead Sea salt (**Table 1**) was added to prepare the required water salinities of 3.0 ppt, 5.5 ppt and 7.0 ppt for the experiment. Continuous aeration was provided by a blower and diffused air stone in each of the rearing tanks while maintaining the oxygen levels in the water at >4 ppm. The fish in the rearing systems were subjected to natural photoperiod while water temperature was maintained at 27±1ºC throughout the experiment using thermostatically controlled heaters (Jagers, Germany). Total ammonia and nitrite were kept at levels below those determined as limits for the sea bream. On day 74 of the experiment, fish were transferred to larger tanks (100-l capacity) for further rearing.

Table 1: Composition of Dead Sea salt (Dead Sea Works Ltd, Israel)

Major elements	Percentage composition
Sodium chloride	98.3
Sulphates	0.5
Calcium	0.3
Magnesium	0.15

Sea bream fingerlings (average weight: 0.4±0.14 g and average length: 2.6± 0.09 cm), produced by Ardag Limited, Eilat, Israel were used for this feeding trials. The three different diets used in the experiment were: Diet 1 "Denis premium 1" feed (Shivuk Raanan, Israel) containing 54% protein and 14.5% fat serving as the control while Diet 2 and Diet 3 were prepared by adding 0.5% and 1.5% Red Sea salt (Red Sea Fish Farm Limited, Eilat), respectively to the "Denis premium 1" feed, which was ground and re-pelleted after adding the salt using a feed pelletizer machine (WLS Loser, Germany). The resulting pellets were oven dried at 60ºC for 6 hours and later stored in air tight containers. Fish in all the tanks were fed ad libitum five times daily by hand. Uneaten feed and excreta were siphoned out daily to minimize water pollution.

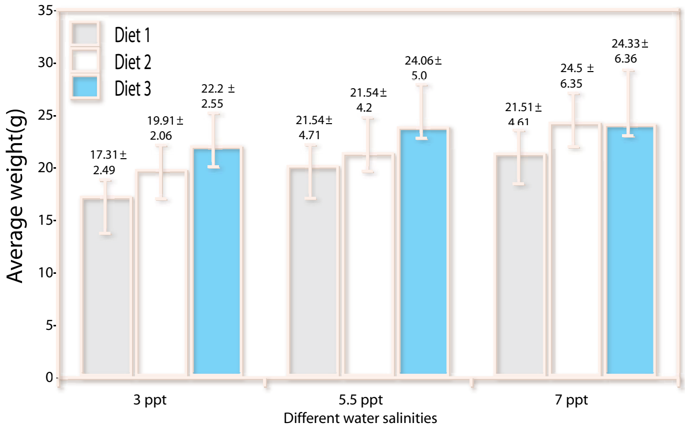

Fig 2: Final weight of gilthead sea bream reared in different salinities fed different salt-enriched diets for 140 days

Although all experimental diets were equally accepted by the fish, those reared in 3.0 ppt salinity and fed the diet containing 1.5% salt grew significantly better ($p<0.05$) than those reared in the same system but fed the control diet. The fish reared in 5.5 ppt and 7.0 ppt water salinities and fed the control or the different salt-enriched diets did not differ significantly. Adding 0.5% sea salt to the diet promoted better survival in 3.0 ppt salinity (42.1%) and 7.0 ppt salinity (50.7%). The specific growth rates (SGR) of fish reared in 5.5 ppt and 7.0 ppt salinities and fed the control diets was lower than when fed salt rich diets, but statistically not significant. In contrast, when reared at 3.0 ppt salinity and fed the diet containing 1.5% salt the SGR was significantly higher than the control diets (**Table 2** and **Fig. 2**).

In a related study, Salman and Eddy (1987) have reported that the food intake and appetite of the rainbow trout were not affected by salt addition to the diet but the addition of salt did enhance the growth of the fish. They also observed a positive linear relationship between the increased level of salt in the diet and in the chloride cell numbers and gill Na+/K+ -ATPase activity. This confirms that fish reared in saltwater are also exposed to additional salt from their food.

However, such quantity of salt is negligible compared to the overall amount of salt from the environment that the fish absorbs through their intestines and body surface. This could explain the trend which the present experiment showed that fish reared in higher salinity achieved higher weight than in lower salinity but not significantly. In another experiment, Gatlin et al (1992) reported that juvenile red drum exhibited greater feed efficiency and significant weight gain when 2% NaCl was added to their diet. In the case of barramundi, adding 4% salt to the diet resulted in significant improvement in their feed conversion ratio (FCR) (Harpaz et al. 2005). In the same experiment, the addition of salt was reported to enhance the activity of the brush border enzymes alkaline phosphatase, lactase, and to some extent, leucine amino peptidase.

The improved enzymatic activity in the fish resulted in better absorption and digestion of food especially in carnivorous fish like the sea bream. In the present study, the SGR of fish reared in 5.5 ppt and 7.0 ppt salinities and fed the control diets was lower than those fed the salt-enriched diets, but statistically not significant. In contrast, fish reared in 3.0 ppt salinity and fed the diet containing 1.5% salt had SGR

Table 2: Growth performance of gilthead sea bream reared in different water salinities for 140 days and fed different salt rich diets

System	Water salinity (ppt)	Tank no.	Types of feed	Average initial weight (g)	Average final weight (g) ± SD	Survival (%)	SGR (%)
1	3	1	Diet 1 (Control)	0.4	17.31 ±2.49^{a}	37.8	2.68^{a}
		2	Diet 2 (0.5% salt)	0.4	19.91± 2.06^{ab}	42.1	2.78^{ab}
		3	Diet 3 (1.5% salt)	0.4	22.20± 2.55^{b}	35.7	2.86^{b}
2	5.5	1	Diet 1 (Control)	0.4	20.30± 4.71	41.2	2.80
		2	Diet 2 (0.5% salt)	0.4	21.54± 4.20	47.7	2.83
		3	Diet 3 (1.5% salt)	0.4	24.06± 5.0	48.9	2.92
3	7	1	Diet 1 (Control)	0.4	21.51± 4.61	43.4	2.83
		2	Diet 2 (0.5% salt)	0.4	24.50± 6.35	50.7	2.92
		3	Diet 3 (1.5% salt)	0.4	24.33± 6.36	49.3	2.92

significantly higher than those fed the control diets. The average survival rate of the fish reared in 3.0 ppt salinity was only 38.53% while the highest average survival rate of 47.8% was attained in fish reared in 7.0 ppt salinity.

From the results of this present experiment, it is evident that long-term exposure of sea bream to low salinity (3.0 ppt) water resulted in growth reduction and low survival rate. The lower the salinity of the test rearing water, the lower was the survival rate. However, the diet containing the highest level of salt (1.5%) could improve the fish growth significantly when reared at lower rearing salinity (3.0 ppt). The results have also indicated that the effect of the salt-enriched diet on the sea bream was more pronounced in the lower salinity water in which the fish were reared. Adding 1.5% sea salt to the diet makes it possible for the fish to grow in water with the lowest tested salinity of 3.0 ppt as well as in highest tested salinity of 7.0 ppt.

Conclusion

From this experiment, it was shown that the use of low salinity water (a mix of brackish water and sea salt) for rearing sea bream resulted in the reduction of growth and increase in mortalities. The experiment indicated that the lower the salinity of the mixed water, the more pronounced was the effect on growth and survival. However, the addition of sea salt to the feed improved the growth and survival rates. The lower the salinity of the rearing water the more effective was the salt-enriched diet on the growth performance. Thus, the findings from this experiment promotes the prospects of euryhaline fish culture in low water salinity opening up a promising future for marine fish production in regions where low saline geothermal water of high quality is available.

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