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Surprising results in rabbitfish diet evaluation

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Soybean meal not a suitable fishmeal replacement for this herbivorous species

The marbled rabbitfish (*Siganus rivulatus*) is a member of a family of tropical algaevorous Indo-Pacific fishes, which successfully invaded the Mediterranean after the opening of the Suez Canal in 1869 and is today endemic to the Eastern Mediterranean (Cyprus, Egypt, Turkey, Lebanon, and Israel). This species is characterized by relatively fast growth and tolerance to high densities in culture, as well as tolerance to wide ranges of salinity and temperature, which makes it a suitable candidate for aquaculture. These factors, in addition to the herbivorous nature of the fish and its demand in the local markets, encouraged research into the species' culture potential.

There is a need for aquaculture of algaevorous fish in order to improve sustainability and green credentials of the industry, and it is imperative to decrease fishmeal in the diets of cultured fish and replace it with a sustainable product with ample protein content such as soybean meal.

Soybean meal is widely used as a partial plant-protein substitute for fishmeal in feeds for a variety of herbivorous/omnivorous species such as catfish, milkfish and tilapia. However, complete substitution of fishmeal with soybean meal is complicated by the fact that soy is deficient in certain essential amino-acids (such as methionine). Also, the presence of anti-nutritional factors in soy can impair nutrient absorption and decrease the nutritive value of the feed, and hence negatively affect growth and

The marbled rabbitfish is an economically valuable species and a suitable candidate for warm water aquaculture in the Eastern Mediterranean.

even health of the animal.

While some of the negative effects of soybean meal on growth can be minimized or avoided through proper processing and amino-acid supplementation, the extent to which it can be used in fish feeds varies from one species to another, even among herbivorous fish.

In our study at the American University of Beirut, we evaluated the use of soybean meal to partially or fully replace fishmeal in the diets of juvenile marbled rabbitfish, by assessing the effects of graded levels of dietary soybean meal on survival, growth and hematological parameters of the fish.

Recirculating system used for the study at American University of Beirut.

Experimental setup

Marbled rabbitfish juveniles were caught off the shore of Ras-Beirut and transported to the aquaculture research facility at the American University of Beirut. Prior to the experimental trials fish were quarantined in an outdoors recirculating system for a month and trained to accept commercial feed. The fish were then size-sorted to a uniform size and a sample of 30 fish was used to calculate initial condition indices from individual body weight and total length. Another sample of 21 fish were frozen whole and stored for whole body composition analysis to serve as initial sample.

The remaining fish were stocked into a closed recirculating system (15 fiberglass tanks measuring 65×64×55cm; L×W×H connected to a bio-filter and a sump tank). Aeration in the system was provided using a regenerative blower and submerged air-stones, temperature and salinity were maintained at levels suitable for optimum growth in the species (27 degrees-C and 35 ppt, respectively) and photoperiod at 14:10 hrs (light:dark). Water quality (dissolved oxygen, ammonia nitrogen, nitrite nitrogen) was maintained at acceptable levels. Fish were stocked at a density of 16 fish per tank, and three tanks were randomly assigned to one of the five prepared diets.

The diets were formulated to contain 40 percent crude protein and 5 percent crude lipid with similar digestible energy (DE; 14 MJ/Kg). Fishmeal was substituted by soybean meal to meet dietary protein proportions equivalent to 25 percent, 50 percent, 75 percent, and 100 percent protein from soybean meal.

The feeding trial lasted eight weeks. Every two weeks the fish in each tank were group-weighed (after a day of fasting) to assess growth. The feed was offered manually at a ration of 6 percent of the largest average body weight and the amount of feed adjusted according to new weight and number of fish in each tank.

At the end of the feeding trial, individual body weight and total length of all fish in the system were measured. A sample of four fish from each tank was used to determine hepatosomatic and viscerosomatic indices (HSI and VSI, respectively) and whole body composition (moisture, protein, lipid and ash proportion in the body). Additionally, four fish from each tank were anaesthetized using a solution of Tricaine-S and blood collected using cardiac puncture to determine red blood cell and white blood cell counts, differential counts of white blood cells, packed cell volume, plasma protein and hemoglobin.

The first author with a juvenile rabbitfish from the study.

Effect on fish growth

Although fish grew in all treatments, growth was affected by the addition of soybean meal in the diet and decreased as more soybean meal was used to substitute fishmeal, with least growth evident in fish offered diets devoid of fishmeal (Figs. 1 and 2). Fish fed diets containing more than 25 percent soybean meal as protein in the diet grew less than fish with no soybean meal in their diet. While breakpoint analysis of the mean body weights among the five treatments shows that soybean meal can constitute approximately 47 percent of the diet before the growth is severely affected, linear regression analysis suggests that growth is negatively correlated with inclusion of soybean meal at any inclusion level. Specific growth rate of fish decreased from 3.85 percent day^{-1} to 3.62 percent day^{-1} when soybean meal was included in the diet (Table 1).

Fig. 1: Growth in average individual body weight (g) over eight weeks of juvenile S. rivulatus, offered diets with increasing inclusion of soybean meal.

Protein utilization and protein efficiency ratio also decrease when more than 50 percent of protein in the diet was of soybean meal origin. Even though marbled rabbitfish is considered to be an herbivorous fish, these results suggest that it is unable to utilize soy protein efficiently, possibly because it feeds on algae in its natural habitat.

Fig. 2: Final body weight (g) as a function of increasing soybean meal inclusion in the diet (g/kg).

Treatment	IBW ± SE	FBW	TL	K	SGR	FE	PER	GPU	S
D1	1.7 ± 0.04	17.2a	10.9a	1.32a	3.85a	0.80a	1.87a	0.32a	97.7
D2	1.9 ± 0.05	16.3ab	10.8a	1.28a	3.62b	0.74b	1.83a	0.33a	83.1
D3	1.8 ± 0.04	15.2b	10.5ab	1.27a	3.61b	0.69b	1.58b	0.28b	84.2

Treatment	IBW ± SE	FBW	TL	K	SGR	FE	PER	GPU	S
D4	1.8 ± 0.10	13.9c	10.2b	1.28a	3.47bc	0.63c	1.59b	0.28b	83.3
D5	1.7 ± 0.07	12.6d	10.1b	1.20b	3.36c	0.56d	1.29c	0.22c	82.2
PSE1	–	0.35	0.12	0.014	0.048	0.015	0.038	0.007	3.39

Effect on whole body composition of fish

Whole body composition of *S. rivulatus* indicates a decrease in tissue lipid when fish were fed soybean meal (Table 2). This could be attributed to lower lipid digestibility caused by anti-nutritional factors present in soybean meal, or high proportion of indigestible carbohydrates in soy which would lower the digestible energy of the feed. Since soybean meal is known to cause morphological changes and inflammation in the gut of the fish (e.g. intestinal enteritis in Atlantic salmon), it is even possible that the health of the fish was affected and more energy was needed to cope with possible inflammation. A slight increase in VSI of fish fed full soybean meal diet could be an indication of a start of such an event.

Treatment	Moisture	Protein	Lipid	Ash
Wild fish	75.30a	14.26b	2.93c	5.54a
D1	69.11c	16.90a	10.30a	3.61b
D2	69.89bc	17.47a	9.17ab	3.70b
D3	70.69b	17.20a	8.45b	3.90b
D4	69.50bc	17.33a	8.86ab	3.73b
D5	70.66b	16.84a	8.13b	3.93b
PSE1	0.389	0.253	0.399	0.111

Health condition of the fish was severely affected when fishmeal was completely removed from the diet as is indicated by Fulton's condition index (K) (Table 1). Although total blood cell counts and blood composition analysis did not suggest stress or health impairment (no significant differences among treatments were observed), it is worth noticing that packed cell volume was considerably lower in fish offered 100 percent soybean meal diets than in fish in other treatments.

Since there were no significant differences in total red blood cell counts among treatments, we suspect a decrease in red blood cell volume as a result of soybean meal inclusion in the diet. Soybean meal also affected liver size in marbled spinefoot as is indicated by a decrease in hepatosomatic index (HSI) (Table 3). Such results can be again attributed to the activity of anti-nutritional factors, which are capable of impairing liver metabolic function and even cause morphological damage.

Treatment	HSI	VSI
D1	2.57a	12.41
D2	2.24b	12.72

Treatment	HSI	VSI
D3	1.81cd	12.33
D4	2.03bc	12.30
D5	1.63d	13.66
PSE1	0.098	0.440

Perspectives

The results from our study clearly suggest that soybean meal is not a viable option when considering fishmeal substitution in the diets for marine herbivore *S. rivulatus*. Possibly, soy protein concentrate might be able to substitute for fishmeal but that remains to be tested. Also, results of the present work are surprising, considering that rabbitfishes are herbivorous and supposedly capable of performing well when offered diets without animal sourced protein. Work continues on trying to formulate a suitable diet that does not contain fishmeal. In the meantime, rabbitfish farms will continue to use traditional marine fish diets for their animals.

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