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Effects of *Ulva rigida* on the Growth, Feed Intake and Body Composition of Common Carp, *Cyprinus carpio* L.

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Abstract: This study is the first trial to evaluate algae meal, *Ulva rigida*, as an inexpensive and locally available feed ingredient in the diet of common carp, *Cyprinus carpio*. Five experimental diets were supplemented with *Ulva* meal at 0, 5, 10, 15 and 20% (C0, U5, U10, U15 and U20, respectively) to investigate the effect of replacement of wheat meal by *Ulva* meal for common carp, *Cyprinus carpio* during a 112-day growth trial. Carp fingerlings, each initially weighing 3.1 g, were stocked into 60 l-glass tanks and were fed to apparent satiation three times daily. Performances of fish fed the test diets were evaluated in terms of survival, final mean weight, percent weight gain, specific growth rate, feed conversion ratio, protein efficiency ratio and body composition. Poorest growth performance was recorded from fish fed the diet with 20% *Ulva* meal supplementation (U20) ($p < 0.05$). Fish group fed with 5% *Ulva* meal inclusion (U5) achieved the best growth performance ($p > 0.05$). Results suggested that the dietary *Ulva* meal inclusion of 5 to 15% replacing wheat meal in carp diets could be acceptable.

Key words: *Ulva rigida*, algae meal, *Cyprinus carpio*, nutrient utilization, body composition growth performance

INTRODUCTION

Nutritionally balanced fish feeds generally contain fishmeal, soy products meal and wheat meal. Investigations for cheaper alternative feedstuffs as protein and energy source for fish diets have become a priority in order to produce low-cost pelleted feeds available for the small-scale farmers both in developed and developing countries. Continuously increasing demand for fish feed in Turkey pressures the consideration of every possible natural resource as a potential ingredient in fish feed.

Algae meals are an alternative plant feedstuffs that are increasingly being used in aqua feeds because of their nutritional quality, lower cost and availability (Mustafa and Nakagawa, 1995; Hassim and Maat-Saat, 1992). *Ulva* sp. belongs to the class *Chlorophyceae* and order *Ulvales*. The specific species of *Ulva rigida* is grown in high mass on the shores of Dardenelles, Turkey. In a study conducted by Wahbeh (1997) shows *Ulva* sp. to be rich in nutritive value; hence, could serve as an alternative fish feed ingredient. The effects of this algae on fish growth, feed efficiency and nutrient utilization have been examined for several species, including black

sea bream, *Acanthopagrus schlegeli* (Nakagawa *et al.*, 1987), snakehead, *Channa striatus* (Hashim and Hassan, 1995), red sea bream, *Pagrus major* (Mustafa *et al.*, 1995) and striped mullet, *Mugil cephalus* (Wassef *et al.*, 2001). The common carp, *Cyprinus carpio* is an omnivorous fish and can utilize feed including plant materials effectively.

This study was aimed to investigate efficiency of *Ulva* sp. as a feed ingredient, on growth, feed intake, nutrient utilization and body composition of common carp.

MATERIALS AND METHODS

Experimental fish, rearing condition and feeding regime:

This evaluation was performed with common carp, *Cyprinus carpio* fingerlings, which were obtained from the fish culture facility of the Institute of Mediterranean Fisheries Research Production and Training (Antalya/Turkey) and transported to Aquarium Units of Faculty of Fisheries, Çanakkale Onsekiz Mart University. They were held under optimal conditions for a period of 2 weeks before beginning the growth trial.

The growth trial lasted for 16 weeks and was carried out in 60 L glass aquariums. Each experimental diet was

fed to triplicate groups of 10 common carp fingerlings with an average initial body weight of 3.0 g. The fish were kept in daily photoperiod of 9 h of light and 15 h dark for the duration 16 weeks time. Feces and pellet residues were discarded by siphoning and 10% of the water in each aquarium was exchanged daily. Water temperature, dissolved oxygen and pH were maintained at 26.0±1.1 °C, 7.4±1.0 and 8.7±0.2 mg L⁻¹, respectively.

Fish were fed to apparent satiation, three times a day at 08:30, 12:30 and 16:30 h. The weight of each fish was measured at the beginning and at the end of the study following a 24 h starvation period allowing gut emptiness.

Preparation of seaweed meal: The *Ulva rigida* seaweed species was collected from the coastal areas of Dardanelles. This species was chosen because of its abundance, distribution and proximity to areas. *Ulva* samples were thoroughly washed with sea water, dried at 40°C for 48 to 72 h and then ground to powder after which a proximate composition of samples was analyzed (Table 1).

Experimental diets: Four experimental *Ulva* meal-based diets were formulated containing 5, 10, 15 and 20% incorporation levels, respectively, by substituting wheat meal on an equal weight basis. A fifth test diet which had no *Ulva* meal supplementation served as the control (C0) diet (Table 2). Diets were formulated to contain 40% total protein and 8% lipid. After thoroughly mixing the dry ingredients, oil and 30% distilled water was added after which the experimental diets were pelleted with a laboratory pelleting machine and dried at 40°C in a fan assisted drying cabinet. Experimental feeds were stored at deep-freezer (-20°C) until used.

Chemical analysis: Proximate composition of feedstuffs, experimental diets and fish body were determined according to AOAC methods (2001): dry matter (DM) after drying in an oven at 105°C until constant weight; crude protein (N = 6.25) by Kjeldahl digestion and distillation after acid digestion; crude lipid by petroleum ether extraction in a Soxhlet apparatus; ash by incineration in a muffle furnace at 550°C for 8 to 12 h.

Data processing and statistical analysis: Fish performance in terms of weight gain (WG, %), feed intake (FI), specific growth rate (SGR), feed conversion ratio (FCR), protein efficiency ratio (PER) apparent net protein utilization (ANPU, %) and nutrient retention was determined using formula in Table 3.

The data were subjected to analysis of variance (ANOVA) and the multiple range test (p<0.05) of Duncan

Table 1: The proximate analyses of *Ulva* meal and wheat meal (%)

Nutrient composition	<i>Ulva</i> meal	Wheat meal
Moisture	11.50	8.00
Protein	8.00	8.63
Lipid	0.15	1.96
Ash	26.40	0.90

Table 2: Ingredients (g kg⁻¹) and nutrient composition (%) of experimental diets

Ingredients	Groups of trial				
	C0	U5	U10	U15	U20
<i>Ulva rigida</i> meal ¹	-	5.0	10.0	15.0	20.0
Fish meal ²	35.0	35.0	35.0	35.0	35.0
Corn gluten ³	10.0	10.0	10.0	10.0	10.0
Soybean meal ⁴	10.0	10.0	10.0	10.0	10.0
Sunflower meal ⁵	5.0	5.0	5.0	5.0	5.0
Wheat meal ⁶	34.0	29.0	24.0	19.0	14.0
Fish oil ⁷	2.0	2.0	2.0	2.0	2.0
Vitamin premix ⁸	2.0	2.0	2.0	2.0	2.0
Mineral premix ⁹	2.0	2.0	2.0	2.0	2.0
Nutrient analysis					
Protein	40.1	40.0	40.0	40.2	40.1
Lipid	8.3	8.2	8.2	8.0	8.1
Ash	10.0	11.5	12.5	12.4	12.5

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using the statistical software package, Statgraphics 7.0 (Manugistics Incorporated, Rockville, MD, USA) (Zar, 2001).

RESULTS

Survival rate of carp (*C. carpio*) remained high, ranging between 84.7 and 94.7% in the experiment without a significant difference (p>0.05) among the treatments (Table 3).

The mean feed intake of fish fed the U20 diet was significantly (p<0.05) lower (below 3% BW) than that of other treatments (C0, U5, U10 and U15) which fed above 3% BW of feed in the experiment (Table 3).

The best growth response was obtained by carp fed on the U5 diet (Table 3). Increase in dietary *Ulva* meal resulted in progressively reduced growth performance, but the final weights attained by carps fed on C, U5, U10 and U15 were not significantly different (p>0.05).

FCR of C and U5 groups were similar, whilst FCR of U5 treatment was 36.4 and 31.8% superior when compared to U10 and U15 groups, respectively. FCR of fish fed with the U20 diet performed poorly.

Dietary protein was efficiently (PER) used by C0 and U5 groups, on the other hand the groups with the highest dietary algae inclusion failed to match this efficiency. Apparent Net Protein Utilization (ANPU) for all treatments ranged from 8.7 to 27.4%. ANPU of U10 and U15 groups were similar, whilst ANPU of fish fed with the U20 diet

Table 3: Growth, feed and nutrient utilization of *C. carpio* fed the experimental diets

Ingredients	Groups of trial (X±SD)				
	C0	U5	U10	U15	U20
Initial mean weight (g)	3.1±0.61	3.1±0.61	3.1±0.61	3.1±0.61	3.1±0.61
Final mean weight (g)	9.2±0.35 ^b	10.0±1.01 ^b	7.9±0.09 ^b	7.4±1.11 ^b	5.1±0.16 ^a
Survival (%)	97.7±5.50	84.7±8.20	88.3±4.95	87.6±8.65	94.0±5.15
Weight gain (%) ¹	194.0±10.53 ^b	221.3±32.60 ^b	155.6±2.71 ^b	150.7±35.77 ^b	64.1±4.87 ^a
Feed intake (%) ²	3.4±0.19 ^{ab}	3.5±0.22 ^b	3.4±0.19 ^{ab}	3.2±0.21 ^{ab}	2.8±0.17 ^a
Feed conversion ratio ³	2.3±0.38 ^a	2.2±0.35 ^a	3.0±0.41 ^a	2.9±0.39 ^a	5.0±0.48 ^b
SGR (%) ⁴	0.96±0.84 ^b	1.03±0.09 ^b	0.84±0.01 ^b	0.80±0.12 ^b	0.44±0.03 ^a
PER (%) ⁵	1.3±0.19 ^b	1.3±0.22 ^b	1.0±0.08 ^{ab}	1.1±0.30 ^{ab}	0.6±0.10 ^a
ANPU (%) ⁶	15.1±0.58 ^a	27.4±0.72 ^d	19.6±0.65 ^b	14.4±0.52 ^b	8.7±0.53 ^a

* Values in each row allocated common superscripts or without superscripts are not significantly different from each other (p>0.05), ¹Weight Gain, WG (%) = [(final weight (g)-initial weight (g))/initial weight (g)] x 100, ²Feed Intake, FI (%) = (daily feed intake (g) x 100)/biomass (g), ³Feed Conversion Rate, FCR = Feed intake (g)/weight gain (g), ⁴Specific Growth Rate; SGR (% day⁻¹) = 100 x [(ln final fish weight)-(ln initial fish weight)]/experimental days, ⁵Apparent Net Protein Utilization, ANPU (%) = [(final body protein (g)-initial body protein (g))/dietary protein consumption (g)] x 100

Table 4: Body composition of *C. carpio* (L.) fed the experimental diets

Carcass component (%)	Groups of trial (X±SD)					
	Initial	C0	U5	U10	U15	U20
Moisture	82.7±2.91	82.2±2.55	79.0±3.20	74.4±2.05	77.2±3.40	83.1±2.18
Protein	11.6±1.10	13.1±0.95	15.5±1.10	16.2±0.61	15.7±1.20	15.2±0.68
Lipid	2.0±0.49	2.7±0.52 ^a	4.0±0.82 ^{ab}	4.6±0.32 ^b	5.1±0.83 ^b	3.5±0.37 ^b
Ash	2.2±0.38	2.1±0.45	3.3±0.73	2.8±0.17	2.3±0.82	2.5±0.18

* Values in each row allocated common superscripts or without superscripts are not significantly different from each other (p>0.05)

performed poorly. The ANPU of fish fed the U5 diet was significantly (p<0.05) superior than that of other treatments during the experiment (Table 3).

Variations in carcass composition of the experimental fish at the beginning and at the end of feeding trial (Table 4) were not significantly different (p>0.05) for ash, protein and moisture content. Final protein content of the fish was increased compared to initial fish in all groups. Both fish fed on control diet and diets included with a variety of *Ulva* meal showed increased lipid content in the final body composition. But this increase was weak for fish fed on the diet supplemented with 20% *Ulva* meal (p<0.05).

DISCUSSION

Natural resources as possible feed ingredients or additives need to be investigated for the development of inexpensive feeds in the world. Macro and micro algae, seaweed and other alternative aquatic plants have been researched and used for decades in aquaculture. *Ulva* meal is one of the most studied and promising source in this sense. In this study, we primarily laid out the possibility of inclusion of *Ulva* meal in common carp fingerlings without affecting their health status negatively.

Earlier reviews detailed that the supplementation of macro and micro algae meals enhanced growth, feed utilization, lipid metabolism, body composition, disease resistance and carcass quality of a variety of fish such as black sea bream, *Acanthopagrus schlegeli*, red sea bream,

Pagrus major, rainbow trout, *Oncorhynchus mykiss* (Mustafa and Nakagawa, 1995). In this experiment the supplementation of *U. rigida* in the range of 5 to 15% to the diet not only improves the growth performance but also the quality of carp as a protein product.

Ten percent *Ulva* meal supplemented feed was consumed by black sea bream weighing 24 g in the study of Nakagawa *et al.* (1987) which resulted in improved physiological condition. A further study (Nakagawa *et al.*, 1993) with black sea bream, initially weighing 56 g each, found that wintering fish for 150 days after feeding for 63 days on diets supplemented with *Ulva* meal up to 10% did not cause a significant loss in body weight and feed efficiency compared to control group. Also the fish groups fed the algae meal supplemented diets utilized accumulated intraperitoneal fat during feeding period more efficiently than control group. Studies conducted with fingerlings displayed better growth by supplementing dietary *Ulva* meal in higher concentration. This could be exemplified with a study where Mustafa *et al.* (1995) compared growth performances of red sea bream fingerlings fed with feeds with three species of algae meal was restricted in terms of its supplementation rate which was only 5%. Fingerlings of 2.1 g consumed these diets where one of the feed was supplemented with *Ulva* meal showed better growth performance than control groups. Moreover, Wassef *et al.* (2001) aimed to determine the optimum level of dietary *Ulva* meal inclusion in mullet fingerlings. They recorded better growth enhancement and higher body protein deposition by feeding mullet fingerlings of 6.4 g with 20% *Ulva* meal supplemented diet

than the groups fed diets with lower *Ulva* meal inclusion. In the present study, addition of *Ulva rigida* meal into diet up to 15% resulted in better growth performance and higher body protein gain concentration for common carp fingerlings comparing to control and groups fed with lower dietary *Ulva* meal levels. Such high concentrations of *Ulva* meal inclusions could also be considered for fingerlings of different fish species.

There have been considerably high numbers of work done on carp species concerning their utilization ability of algae meal sorts, aquatic and terrestrial macrophytes in raw, fermented, ensilaged, autoclaved or any other form in the diet. Ray and Das (1992) successfully incorporated composted *Salvinia cuculata* until 20% dietary rate without compromising the growth performance. Bairagi *et al.* (2002) concluded that feeding rohu (*Labeo rohita* Ham.) fingerlings, a species of Cyprinidae family, with diet incorporating up to 30% fermented duckweed (*Lemna polyrhiza*) leaf gave better growth response, feed efficiency, higher protein and lipid gain compared to 10% raw duckweed inclusion level. Contrastingly, another species which is a member of Cyprinidae, catla (*Catla catla*) fingerlings were fed (Harish Kumar *et al.*, 2004) with increasing amounts of dietary *Spirogyra* sp. meal, belonging to the class *Chlorophyceae*, showed worse growth performance even at the lowest dietary inclusion level. At the end of the day, the success of usefulness of increased dietary inclusion algae species in to diet depends on the fish species, algae species and the form in which the algae is delivered.

This study is a first attempt to evaluate the inclusion of *Ulva* meal in common carp diet. We assume that product quality of fish in terms of higher protein accumulation in weight gain could capture attention of fish producers by inclusion of *Ulva* meal to significantly high dietary concentrations like up to 15% in fingerlings. Hence, the present study suggests that *U. rigida* is a rising candidate in terms of feed additive which could be used in common carp feed.

Innovation of cost-effective technologies for cultivation of *U. rigida* would boost the use of this algae as a fish feed additive. Further experiments are required with a range in fish body weight and added investigations of digestibility of dietary nutrients and lipid metabolism.

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