Effect of Algae as a Feed Additive on Growth Performance in Red Sea Bream, *Pagrus major*

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ABSTRACT

Algae as a feed additive improved physiological condition including protein assimilation, lipid metabolism, liver function, stress responses, disease resistance and carcass quality of cultured fish. Effects on the growth performance, feed efficiency, and results of related biochemical analyses of red sea bream fed on algae-supplemented diet are discussed. The algae *Ascophyllum nodosum*, *Porphyra yezoensis*, *Spirulina* sp. and *Ulva pertusa* were supplemented to the zero year and one-year-old red sea bream diet at 3-5%. Feeding algal meal as a feed additive elevated growth rate, feed efficiency, protein efficiency ratio and muscle protein deposition. An increase in RNA/DNA ratio and decrease in acid proteinase activity were observed in algae-fed fish. Among the algae tested in these experiments, the effects were pronounced with *Spirulina*. The results confirmed desirability for algae as a feed additive in growth performance of red sea bream.

INTRODUCTION

The addition of a small amount of algal meal to the fish diet can produce considerable effects on the growth, feed utilization, physiological condition, stress response, disease resistance, body constituents and carcass quality of cultured fish.[1] The study of fish nutrition have progressed considerably with regard to elevation of growth rate and feed efficiency. With regard to the effects of dietary algae on growth in fish, a 5% supplement of *Chlorella*, *Spirulina* and *Ulva* meal resulted in a higher body weight gain in the nibbler *Girella punctata*.[2] In addition, elevation of feed efficiency and weight gain have been reported in red sea bream fed with *Undaria pinnatifida* and *Ascophyllum nodosum* as a feed additive.[3]

This study further confirmed the effect of algae meal as a feed additive on growth performance in red sea bream in two sets of experiments. We investigated the effects of a brown alga *Ascophyllum*, a red alga *Porphyra*, a green alga *Ulva* and a blue-green micro-alga *Spirulina* since these have been studied most extensively with respect to use as dietary additives in fish feed.

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MATERIALS AND METHODS

Zero year and one-year-old red sea bream were used in the experiments after a two-week acclimatization period in the Fisheries Laboratory of Hiroshima University. Four species of algae, *Ascophyllum nodosum*, *Porphyra yezoensis*, *Ulva pertusa*, and *Spirulina* sp., were supplemented to the basal diet shown in Table 1 at 3 or 5%. Water temperature and salinity ranged from 21-18°C and 26-34ppt, respectively and water flow rate was 10L/min.

Feed efficiency (FE), protein efficiency ratio (PER), daily growth rate (GR), muscle protein deposition (MPD) and protein sparing effect (PSE) were calculated from the following equations :

FE (%) = weight gain/diet fed × 100

PER = weight gain/protein fed

GR (%) = individual weight gain/days × 100

MPD (%) = muscle protein gain/protein fed × 100

PSE (%) = 100 − (protein required for 100g weight gain in experimental fish/protein required for 100g weight gain in control fish × 100)

Biological parameters were measured from 10 to 30 fish in each group. Muscle and liver were dissected from 5 to 10 fish from each treatment group. For determination of nucleic acid and proteinase activity, dorsal white muscle was frozen in liquid nitrogen and stored at −80°C. RNA and DNA were measured according to Munro and Fleck.[4] Protein was determined by Folin reagent[5] and acid proteinase activity was measured according to Makinodan et al.[6] Data were analyzed by Duncan's multiple range test.

RESULTS

Experiment I. Zero year red sea bream with an average initial body weight of 2g were reared for 41 days in duplicate one ton plastic tanks each containing 200 fish. A moist-type feed, as shown in Table 1 was fed to satiation three to four times a day (7 : 00, 10 : 00, 14 : 00 and 17 : 00). For the experimental diets, 5% of the ingredients were replaced by *Ascophyllum nodosum*, *Porphyra yezoensis* or *Ulva pertusa* algal meal. Crude protein content of the experimental diet ranged between 38.5 and 39.3%.

The effect of algae supplementation was appeared to be dependent on the amount of diet given. Feeding with algae increased body weight, total biomass and daily growth rate (Table 2). The feed efficiency, protein efficiency ratio and muscle protein deposition tended to be improved by feeding the algal-supplemented diet. The effects were high in the *Porphyra*-fed group followed by the groups fed with *Ascophyllum* and *Ulva*. The muscle ratio, hepatosomatic index and IPF ratio were increased by *Porphyra*

Table 1. Basal diet composition for red sea bream

	Experiment 1	Experiment 2
Ingradients (%)		
Krill (frozen)	17.0	23.8
Sand lance (frozen)	33.0	23.7
Composed diet	50.0	47.5
Cellulose		3.0
Vitamin mixture		1.0
Mineral mixture		1.0
Proximate composition (%)		
Moisture	42.7	46.1
Crude protein	39.3	30.4
Lipid	4.0	4.0
Ash	6.4	6.8

In experimental diets, 5% (Expt. 1) and 3% (Expt. 2) algae were supplemented to the basal diet.

Table 2. Effects of feeding algae on growth performance and feed utilization in red sea bream

	Experiment 1				Experiment 2		
	C	A	P	U	C	P	S
Total diet given (kg)	3.56^{a}	3.78^{b}	3.85^{c}	3.80^{b}	14.27	14.40	14.43
Survival (%)	77.8	84.0	87.8	84.3	100	98	100
Biomass increase (g)	1595^{a}	1975^{ab}	2281^{b}	1816^{ab}	3832^{a}	4269^{b}	4495^{b}
Weight gain (g/fish)	10.9^{a}	12.1^{ab}	13.3^{b}	11.2^{ab}	153^{a}	175^{b}	180^{b}
Daily growth rate (%)	4.38^{a}	4.61^{ab}	4.88^{b}	4.47^{a}	2.49^{a}	2.90^{b}	2.90^{b}
Feed efficiency (%)	51.5	56.8	62.3	52.4	26.9^{a}	29.8^{b}	31.2^{b}
Protein efficiency ratio	1.31	1.48	1.60	1.36	0.88^{a}	0.95^{ab}	0.98^{b}
Muscle protein deposition (%)	9.3	10.7	12.5	9.8	11.7	13.5	13.1
Protein sparing (%)	—	14.9	25.3	6.9	—	9.6	10.5

Each value is the mean of duplicate experiments. Different superscripts on the same row in each experiment indicate significant differences ($p<0.05$).
C : Control ; A : *Ascophyllum* ; P : *Porphyra* ; S : *Spirulina* ; U : *Ulva*.

supplementation (Table 3). Table 4 shows the muscle constituents of the fish. The RNA/DNA ratio as an indicator of protein synthesis and somatic growth was elevated by the algae. While DNA content was not different, protein/DNA ratio as an index of protein concentration per cell of the fish fed *Porphyra* and *Ascophyllum* increased considerably.

Experiment II. One-year-old red sea bream, 60g in initial average body weight, were reared for 103 days in duplicate one ton plastic tanks containing 25 fish in per treatment group. Cellulose in the basal diet was replaced by two types of algal meal, *Porphyra yezoensis* and *Spirulina* sp., at 3%. The fish were satiated with the moist diet twice daily (7 : 00 and 17 : 00).

Both *Porphyra* and *Spirulina* meal in the moist diet produced significant enhancement of growth and feed utilization. Biomass production, weight gain, daily growth rate, feed efficiency and protein efficiency ratio were elevated significantly by feeding with the algae (Table 2). Muscle ratio were considerably elevated by feeding with both *Porphyra* and *Spirulina*, but the other parameters were not influenced by the algal supplement (Table 3). While the RNA/DNA ratio and protein/DNA ratio were not different among the groups, the muscle acid proteinase activity was significantly low in the algae fed fish (Table 4).

Table 3. Effects of feeding algae on biological parameters in red sea bream

	Experiment 1				Experiment 2		
	C	A	P	U	C	P	S
Mean body weight (g)	13.2^{a}	14.8^{ab}	17.2^{b}	15.8^{b}	214^{a}	233^{b}	241^{b}
Muscle ratio (%)	35.2^{a}	35.3^{a}	37.4^{b}	35.2^{a}	46.6^{a}	48.8^{b}	48.3^{b}
Hepatosomatic index (%)	1.12^{a}	1.07^{a}	1.51^{b}	1.25^{a}	2.10	2.26	2.02
Intraperitoneal fat body ratio (%)	0.23^{a}	0.26^{a}	0.51^{b}	0.51^{b}	2.17	2.44	2.11

Each value is the mean of duplicate experiments. Different superscripts on the same row in each experiment indicate significant differences ($p<0.05$).
Dietary groups, see Table 2.

Table 4. Effects of feeding algae on muscle constituents in red sea bream

	Experiment 1				Experiment 2		
	C	A	P	U	C	P	S
Protein (mg/g)	175	175	178	172	168	174	173
RNA/DNA	3.14^{a}	3.72^{b}	4.17^{c}	3.25^{a}	2.09	2.12	2.47
Protein/DNA	4.47^{a}	5.04^{b}	5.03^{b}	4.30^{a}	7.65	8.11	8.50
Acid proteinase*	NA	NA	NA	NA	15.5^{a}	8.3^{b}	7.3^{b}

Each value is the mean of 5 determinations. Different superscripts on the same row in each experiment indicate significant differences ($p<0.05$).
Dietary groups, see Table 2.
*Activity expressed as nanomoles of tyrosine released per mg protein per hour.
NA : not analysed.

DISCUSSION

Algal supplementation in moist type feed has been found to enhance growth and feed efficiency in fish. Among the algae tested in these experiments, the effects were most pronounced by feeding *Spirulina*. The effects of dietary algae on growth performance have been reported in red sea bream[3,7,8] and Japanese flounder *Paralichthys olivaceus*.[9] According to RNA/DNA ratio, protein/DNA ratio and acid proteinase activity, feeding with the algae-supplemented diet influenced protein metabolism, cellular growth and protein deposition. The growth performance in the algae-fed fish might reflect these factors.[10] Dietary algae contributed to the absorption of dietary carbohydrate and protein, and stimulated assimilation

of nutrients into body constituents.[11)]

As biochemical constituents and digestibility differ among algae,[12)] the effects of dietary algae apparently varied with the species of both algae and fish.[13,14)] The algae contributed to the elevation and improvement of growth, feed utilization, physiological activity, stress responses, starvation tolerance, disease resistance and carcass quality of cultured fish.[1,15)] Analysis of feeding behavior demonstrated that many fish, even those which are carnivorous, ingest algae as a food source. Thus, the use of algae as a feed additive would be advantageous for the efficacious utilization of artificial diets in cultured fish.

The practical use of algae in fish diet is on the increase in Japan. About 1% algal meal now generally applied to commercial fish feed. The advantageous effects of dietary algae are assumed to be due to dietary fiber, carotenoids, chemical feeding attractants, vitamin source, synergistic effect with vitamins, minerals, protection against vitamin degradation and a binder for feed preparation. Nakajima[16)] reported the efficacy of algal dimethyl-β-propiothetin in increasing growth and thrust power of fish. *Ulva*-extract was reported to contribute to improvement in the physiological condition of fish suggesting effects derived from multiple algal substances.[17)] However, the mechanisms involved in the efficacy of algae as a feed additive are not clear, so further studies are needed.

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