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Effects of Different Protein and Lipid Levels in Practical Diets for Yellowtail Snapper, *Ocyurus chrysurus* (Bloch, 1971)

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Abstract: Yellowtail snapper *Ocyurus chrysurus* has great potential as a production fish in aquaculture, yet there is very limited information on its nutritional requirements. To establish baseline data, two trials were conducted to evaluate the effects of dietary protein and lipid levels in practical diets on growth and protein retention. The first trial, conducted over 14 weeks, used a series of diets with varying levels of protein (36%, 40%, and 44%) and lipids (6%, 10%, and 14%). The second trial, conducted for 10 weeks, used a series of diets with 36% protein and scaled lipid levels (7%, 10%, 13%, and 16%). Additionally, a commercial reference diet with 44% protein and 12% lipids was included. Growth performance and feed utilization parameters for Trial 1 indicated that the yellowtail snapper were able to effectively utilize practical diets containing 36% protein and 10% lipids, which produced the highest apparent net protein retention (ANPR; %) and survival. No significant differences were found in growth performance metrics, though there were numerical differences in final weight, weight gain, and survival. Similarly, in Trial 2, most growth metrics did not show significant differences. There were variations in weight gain, feed offered, and ANPR, with the highest performance observed in the fish given feed with 13% lipids. Based on the growth performance and ANPR values across these trials, we recommend 36% protein and dietary lipid levels of 7–13%, which are lower than the currently used commercial diets for marine finfish. The data gathered from the current study may be helpful for nutritionists in formulating feed to include more sustainable and cheaper feedstuffs and promote sustainable yellowtail snapper aquaculture production.



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1. Introduction

Mariculture contributes 36% of total economic value generated by aquaculture and 34% of the total volume of aquaculture production. While farmed fish represent a smaller portion of overall marine finfish production as compared to capture fisheries, for the species that are farmed, cultured fish dominate the seafood market. Emerging opportunities to increase mariculture production (e.g., offshore aquaculture [1,2]), and increasing recognition of the potentially lower environmental impacts of mariculture compared with land-based animal products [3,4]), suggest that mariculture holds significant promise for providing sustainable and nutritious food sources to help meet growing protein demand.

Dietary protein is the most expensive component in fish diets [5,6]. Given that protein makes up over 40% of commercial yellowtail diets, optimizing protein levels is crucial for enhancing profitability. These levels of protein in marine fish diets are viewed as crucial

to obtain amino acids for protein synthesis and to meet energy needs [7–9]. Aside from increasing feed costs, excessively high protein feeds can also result in poor fish growth and feed utilization [10,11]. Excesses of dietary protein also increase nitrogenous excretion into the rearing water, resulting in pollution and higher environmental costs [12,13]. Quite often in high protein diets, lipid levels are increased to provide a source of non-protein energy, sparing protein degradation. However, high lipids do not work for all species. For example, digestibility was reduced in cobia (*Rachycentron canadum*) when fed with increasing dietary lipid levels which resulted in excess lipid deposition in the tissues [14]. As there are interactions between these nutrients, studies on optimizing protein and lipid levels must be conducted for diet formulation development. Studies focusing on determining protein and lipid levels promote sustainable cultivation for promising fish species that are good candidates for aquaculture, thereby diversifying the possible species options for food.

Interest has grown in the possibility of culturing the yellowtail snapper, *Ocyurus chrysurus*. This species is distributed in the western Atlantic from North Carolina to southeastern Brazil but is most abundant in the Bahamas, south of Florida, and in the Caribbean. Fishermen seek this fish as a sport fish and regard it as a good quality food fish. Commercially, the species is caught by hook and line, baited trap, trammel and gill nets, trawl, and beach seine. In the U.S. Virgin Islands and Puerto Rico, the species is the most prized snapper [15]. Yellowtail snapper is an important fisheries resource where they occur, with high production in northeast Brazil [16], averaging 1683 tons in 2001 [17] and accounting for 16% of the total fish biomass captured between 1986 and 1989 [18] in southeast Brazil. Biological research on yellowtail snapper has not been extensive and has immense potential to support a promising mariculture industry.

Nutritional information on snapper species is quite limited. There are a few studies on the use of alternative protein and lipid sources to evaluate digestibility and growth performance of snapper species. Australian snapper were studied in this way, using a large range of protein and lipid sources and levels [19–22]. Research with red snapper utilized different protein sources for fishmeal replacement, the use of attractants, and taurine removal [23]. Overall, these studies demonstrated wide acceptability of snapper species to different protein sources.

Presently, there is very limited information on nutrient recommendations for snapper in general and almost none on the yellow tail snapper. To promote enhanced production and culture of yellowtail snapper, basic data on the dietary requirements need to be developed. This study was designed to evaluate nutritional responses of yellowtail snapper using practical diets containing varying levels of protein and/or lipids.

2. Materials and Methods

2.1. Experimental Design and Diets

Two growth trials were conducted to determine basic optimal protein and lipid levels for practical diets for yellowtail snapper (Table 1). For the first trial, five diets were formulated to cover the typical range of protein and lipids that are used in marine fish diets. This included three diets with 36, 40, and 44% protein with 10% lipids (36:10, 40:10, and 44:10, respectively). An additional two diets were formulated with 44% protein and 6 and 14% lipids, thus allowing three diets with 44% protein and 6, 10, and 14% lipids (44:6, 44:10 (from the first set), and 44:14). Based on results of the first trial, the second series of diets was formulated to contain 36% protein and 7, 10, 13, and 16% lipids. A commercial diet, Otohime EP3 (Reed Mariculture, Campbell, CA, USA) was used as a commercial reference for the second series of diets.

Table 1. Diet formulation and proximate analysis of practical yellowtail snapper diets with varying protein and lipid levels for both trials. Analysis was performed by MidWest Laboratories, Inc., Omaha, NE, USA.

Ingredient	Trial 1					Trial 2				
	Protein	36	40	44	44	44	36	36	36	36
	Lipids	10	10	10	6	14	7	10	13	16
Menhaden fishmeal ¹		26.70	30.00	35.10	35.10	35.10	26.70	26.70	26.70	26.70
Soybean meal ²		17.44	19.60	21.56	21.56	21.56	18.00	18.00	18.00	18.00
Corn protein concentrate ³		8.90	10.00	11.00	11.00	11.00	8.60	8.60	8.60	8.60
Menhaden fish oil ⁴		6.80	5.50	5.50	2.00	9.80	3.30	6.35	9.30	12.30
Lecithin (soy) ⁵		1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00
Corn starch ⁶		10.11	5.85	4.79	8.29	0.49	13.35	10.30	7.35	4.35
Wheat flour ⁷		26.00	26.00	19.00	19.00	19.00	26.00	26.00	26.00	26.00
Trace mineral premix ⁸		0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Vitamin premix ⁹		0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50
Choline chloride ⁶		0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20
Rovimix Stay-C ¹⁰		0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10
CaP-dibasic ⁶		1.00	0.00	0.00	0.00	0.00	1.00	1.00	1.00	1.00
Taurine ¹¹		1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00
Proximate composition (g/100 g as is) ¹²										
Protein (crude) %		38	41.6	45.1	45.3	45.1	38	38.3	37.7	37.7
Moisture %		8.84	8.51	8.53	10	9.73	5.33	4.92	4.99	4.54
Fat %		9.82	8.98	9.42	6.43	12.5	7.1	9.8	12.7	15.1
Ash %		91.16	91.49	91.47	90	90.27	9.42	9.31	9.5	9.33
Sulfur %		0.76	0.7	0.79	0.82	0.8	0.74	0.71	0.76	0.69
Phosphorus %		1.45	1.24	1.35	1.42	1.39	1.56	1.48	1.59	1.5
Potassium %		0.83	0.78	0.88	0.89	0.9	0.8	0.76	0.82	0.78
Magnesium %		0.16	0.15	0.16	0.16	0.16	0.16	0.15	0.16	0.15
Calcium %		2.06	1.69	1.91	2	2.06	2.27	2.14	2.25	2.24
Sodium %		0.29	0.28	0.34	0.34	0.35	0.29	0.28	0.3	0.28
Iron %		278	213	294	287	299	240	222	243	224
Manganese %		99.3	46.3	66.4	63.7	66.8	59.4	69.6	63.3	58.2
Copper %		7.3	4.9	6.2	6.3	6	5.6	5.2	6	5.4
Zinc %		201	94.9	170	163	164	150	132	147	112

¹ Special SelectTM, Omega Protein Inc., Houston, TX, USA. ² Solvent Extracted Soybean Meal, De-hulled solvent-extracted soybean meal, Bunge Limited, Decatur, AL, USA. ³ Emphyreal 75 TM Cargill Corn Milling, Cargill Inc., Blair, NE, USA. ⁴ Omega Protein Inc., Reedville, VA, USA. ⁵ The Solae Company, St. Louis, MO, USA. ⁶ MP Biomedicals Inc., Solon, OH, USA. ⁷ Bobs Red Mill Natural Foods, Milwaukie, OR, USA. ⁸ Trace mineral premix (g/100 g premix): cobalt chloride 0.004, cupric sulphate pentahydrate 0.250, ferrous sulphate 4.0, magnesium sulphate anhydrous 13.862, monohydrate 0.650, potassium iodide 0.067, sodium selenite 0.010, zinc sulphate heptahydrate 13.193, filler 67.964. ⁹ Vitamin premix (g/kg premix): Thiamin HCl 0.751, riboflavin 4.505, pyridoxine HCl 1.502, D-Pantothenic acid hemicalcium salt 7.508, nicotinic acid 7.508, biotin 0.075, folic acid 0.270, vitamin B12 0.003, inositol 7.508, menadione 3.003, vitamin A acetate (500,000 IU/g) 0.300, vitamin D3 (1,000,000 U/g) 0.60, DL- α -tocopheryl acetate (250/ IU g-) 12.012, α -cellulose 804.847. ¹⁰ Stay C®, (L-ascorbyl-2-polyphosphate 35% Active C), Roche Vitamins Inc., Parsippany, NJ, USA. ¹¹ TCI (Tokyo Chemical Industry), Portland, OR, USA. ¹² Analysis was performed by MidWest Laboratories, Inc., Omaha, NE, USA.

All diets were formulated in accordance with the nutritional requirements of marine fish [24] and applied to yellowtail snapper. The experimental diets were prepared at the Aquatic Animal Nutrition Laboratory at the School of Fisheries, Aquaculture, and Aquatic Sciences, Auburn University (Auburn, AL, USA), using standard procedures for fish feeds. The dry ingredients and oil were weighed and mixed in a food mixer (Hobart Corporation, Troy, OH, USA) for 15 min. Subsequently, boiling water was then blended into the mixture to obtain a consistency appropriate for pelleting. Diets were pressure-pelleted using a meat grinder with a 3 mm die. The moist pellets were then placed into a forced air oven (VWR Scientific E191047, Radnor, PA, USA) (<45 °C) overnight and were crumbled, packed, and stored in sealed bags in a freezer (−20 °C) after drying. Proximate composition analysis for all experimental diets were analyzed at MidWest Laboratories, Inc., Omaha, NE, USA (Table 1).

2.2. Growth Trial

Both growth trials were conducted at E.W. Shell Fisheries Center at Auburn University, Auburn, Alabama in an indoor recirculation system consisting of culture tanks, reservoir, fluidized biological filter, bead filter, circulation pump and supplemental aeration. For Trial 1, fifteen 730 L polyethylene circular tanks connected to a common reservoir tank (1600 L) were used, with thirty juvenile yellowtail snapper that were batch-sorted to uniform size, group weighed, and stocked into each tank. For trial two, twenty 83 L glass rectangular tanks connected to a common reservoir tank (800 L) were stocked with ten juvenile yellowtail snappers per tank, following the described protocol. Yellowtail snapper for the experiments were obtained from the University of Miami Experimental Hatchery (UMEH) via the Whitney Laboratory for Marine Bioscience at University of Florida.

For each growth trial, three replicate groups of fish per dietary treatment were randomly assigned the test diets. The fish were offered a fixed ration that was divided into two equal feedings each day. Fish were counted and weighed every other week to adjust the daily feed ration, which was calculated based on percentage body weight which took into account the expected growth and the observed feed response. During the weighing process, fish were dipped in a solution of chloroquine phosphate (MP Biomedicals, Solon, OH, USA) at a concentration of 60 mg/L for around 2 min followed by a 10–15 s dip in dechlorinated freshwater. This treatment was used to reduce the likelihood of amyloodinium (*Amyloodinium ocellatum*) infections. At the end of the 14-week growth trial, fish were counted and group weighed by replicate tank to determine mean final biomass, final weight, survival, percent weight gain, feed conversion ratio (FCR), and thermal-unit growth coefficient (TGC). Fish were euthanized and packed in sealed bags and stored in a freezer ($-20\text{ }^{\circ}\text{C}$) for proximate analysis.

2.3. Water Analysis

Dissolved oxygen was maintained near saturation using air stones in each culture tank and the sump tank each connected via a common air distribution system and a regenerative blower. During the trial, dissolved oxygen, salinity, and water temperature were measured twice daily using a YSI-55 multiparameter instrument (YSI corporation, Yellow Springs, OH, USA), and total ammonia N (TAN) and nitrite-N were measured twice per week using a YSI 9300 photometer (YSI, Yellow Springs, OH, USA). The pH of the water was measured twice weekly during the experimental period using a pHTestr30 (Oakton Instrument, Vernon Hills, IL, USA).

During the growth period for Trial 1, DO, temperature, salinity, pH, total ammonia nitrogen (TAN), and nitrite were maintained within acceptable ranges for marine fish culture at 6.51 ± 0.06 mg/L, 26.34 ± 0.1 °C, 28.32 ± 0.16 g/L, 7.83 ± 0.03 , 0.45 ± 0.06 mg/L, and 0.97 ± 0.19 mg/L, respectively, while for Trial 2, DO, temperature, salinity, pH, total ammonia nitrogen (TAN), and nitrite were maintained as follows: 7.24 ± 0.1 mg/L, 27.15 ± 0.11 °C, 23.75 ± 0.33 g/L, 7.89 ± 0.06 , 0.47 ± 0.07 mg/L, and 0.75 ± 0.17 mg/L, respectively.

2.4. Statistical Analysis

All data were analyzed using SAS (V9.4, SAS Institute, Cary, NC, USA). Growth indices of fish, including final weight, weight gain, thermal growth coefficient (TGC), and survival rates, were analyzed using one-way analysis of variance (ANOVA) to determine significant differences ($p < 0.05$) among treatments. This was followed by Tukey's multiple comparison test to perform pairwise comparisons between treatment means.

Proximate composition measurements, including moisture, crude protein, fat, and ash were analyzed in the same way. Data are presented as mean \pm standard error (SE).

3. Results

For Trial 1, no significant differences were observed for all growth performance parameters (Table 2). For fish offered diets with various protein levels (36–44%) with 10% lipids, final weight (14.45 to 15.4 g), weight gain (12.34 g to 13.37 g or 583% to 658%), and

TGC (0.0445 to 0.0473) were numerically lower for fish offered the 36% protein diet and higher for the 40% protein diet, respectively. Final biomass (390.77 to 396.83 g) and survival (84.4% to 87.8%) were reversed as fish offered the 40% protein diet were numerically smaller than those offered the 44% protein diet, respectively. For fish offered diets with various lipid levels (6–14%) with 44% protein, final weight (13.2 to 14.85 g), weight gain (11.24 g to 12.84 g or 572% to 648%), TGC (0.043 to 0.0463), final biomass (317.1 g to 400.67 g), and survival (80% to 90%) were numerically lower for fish offered the 6% lipid diet and higher for the 14% lipid diet, respectively.

Table 2. Response of juvenile yellowtail snapper (mean initial weight; 2.03 ± 0.06 g) fed diets containing various protein and lipid levels within a 14-week period for Trial 1. Values represent the means of three replicates.

Protein:Lipids	Final Biomass (g)	Final Weight (g)	Weight Gain (g)	Weight Gain (%)	Total Dry Feed (g)	FCR ¹	Survival (%)	TGC ²	ANPR ³ (%)
36:10	400.17	14.45	12.34	583.64	31.87	2.61	92.22	0.0445	18.94
40:10	390.77	15.40	13.37	658.47	32.51	2.43	84.44	0.0473	18.89
44:10	396.83	15.07	12.97	618.72	32.34	2.50	87.78	0.0461	16.74
PSE ⁴	13.21	0.44	0.43	19.81	0.76	0.05	1.58	0.000898	0.44
<i>p</i> -value ⁵	0.9678	0.725	0.6736	0.3464	0.9528	0.4612	0.1199	0.5045	0.0567
44:6	317.10	13.20	11.24	572.07	28.20	2.51	80.00	0.0430	18.49
44:10	396.83	15.07	12.97	618.72	32.34	2.50	87.78	0.0461	16.74
44:14	400.67	14.85	12.84	648.02	32.37	2.56	90.00	0.0463	16.44
PSE	18.77	0.49	0.49	28.38	0.89	0.06	2.34	0.00	0.57
<i>p</i> -value ⁵	0.1062	0.2529	0.3105	0.6103	0.0624	0.9326	0.1965	0.4799	0.313

¹ FCR = Feed conversion ratio = feed offered/(final weight – initial weight). ² TGC = Thermal-unit growth coefficient. ³ ANPR = Apparent net protein retention. ⁴ PSE = Pooled standard error. ⁵ One-way analysis of variance (ANOVA) was used to determine significant differences (*p* < 0.05) between treatment means.

No significant differences were observed for all proximate composition parameters (Table 3) such as moisture, crude protein, fat, and ash for all yellowtail snapper fed experimental diets with varying protein and lipid levels.

Table 3. Whole-body composition (on wet weight basis) of yellowtail snapper fed diets with varying protein and lipid levels for Trial 1. One-way analysis of variance (ANOVA) was used to determine significant differences (*p* < 0.05). Analysis was performed by MidWest Laboratories, Inc., Omaha, NE, USA.

Protein:Lipids	Moisture %	Protein (Crude) %	Fat %	Ash %
36:10	69.00	18.30	7.96	4.52
40:10	68.17	18.67	6.69	4.85
44:10	68.50	18.43	8.21	4.52
PSE	0.22	0.31	0.32	0.14
<i>p</i> -value	0.3359	0.9116	0.0896	0.6153
44:6	69.43	20.27	6.05	4.35
44:10	68.50	18.43	8.21	4.52
44:14	68.00	18.43	9.09	4.29
PSE ¹	0.31	0.59	0.58	0.23
<i>p</i> -value ²	0.148	0.3983	0.006	0.9347

¹ PSE = Pooled standard error. ² One-way analysis of variance (ANOVA) was used to determine significant differences (*p* < 0.05).

For Trial 2, no significant differences (Table 4) were observed for all growth parameters, except apparent net protein retention (ANPR; %). The highest weight gain was observed in yellowtail snapper fed the 13% lipid diet and the lowest weight gain was observed in those fed with the 16% lipid diet. For total dry feed offered per fish, the highest values were observed for yellowtail snapper fed commercial feed having 48% protein and 10% lipids, followed by fish fed the 13% lipid diet, and the fish that had the least amount of feed offered were those fed with the 16% lipid diet. For thermal growth coefficient (TGC),

the highest values were demonstrated by fish fed the commercial feed, while among the experimental diets, yellowtail snapper fed the 13% lipid diet had the highest TGC and fish fed the 16% lipid diet had the lowest TGC. Yellowtail snapper fed commercial feed had the most desirable FCR, while among the experimental diets, fish fed the 7% lipid diet and 13% lipid diet had the most desirable FCR, and fish fed the 16% lipid diet had the least desirable FCR. Significant differences were observed in the ANPR of yellowtail snapper. Fish fed the 7% lipid diet had the highest ANPR values, as opposed to those fed the 16% lipid diet and commercial feed, which had the lowest. Significant differences (Table 5) were observed for moisture and fat for whole-body composition of yellowtail snapper when fed experimental diets.

Table 4. Response of juvenile yellowtail snapper (mean initial weight = 3.40 ± 0.06 g) fed diets containing 36% protein with varying lipid levels within a 10-week period for Trial 2. Values represent means of three replicates.

Dietary Lipids	Final Biomass (g)	Final Weight (g)	Weight Gain (g)	Weight Gain (%)	Total Dry Feed (g)	FCR ¹	FCE ²	Survival (%)	TGC ³	ANPR ⁴ (%)
7	161.28	18.81	7.44	446.30	19.42	2.62	78.88	85.00	0.03764	18.65 ^{a,b}
10	151.08	18.79	6.89	455.03	18.83	2.76	81.67	80.00	0.03573	16.65 ^b
13	177.20	20.07	7.69	483.29	20.09	2.62	82.76	87.50	0.03850	18.62 ^a
16	136.28	15.90	6.30	392.79	17.62	2.84	71.56	85.00	0.03405	16.54 ^{a,b}
PSE ⁵	8.84	0.65	0.26	15.73	0.44	0.05	1.92	2.23	0.00	0.14
<i>p</i> -value ⁶	0.4535	0.128	0.244	0.2354	0.2486	0.3569	0.1548	0.7273	0.3441	0.0254
Pro48:Lip10 ⁷	202.70	20.90	9.37	499.62	22.38 ^a	2.40	77.46	97.50	0.04396	15.42

¹ FCR = Feed conversion ratio = feed offered / (final weight – initial weight). ² FCE = Feed conversion efficiency = (final weight – initial weight) / feed offered. ³ TGC = Thermal-unit growth coefficient. ⁴ ANPR = Apparent net protein retention. ⁵ PSE = Pooled standard error. ⁶ One-way analysis of variance (ANOVA) was used to determine significant differences (*p* < 0.05). Tukey’s multiple comparison test was used to determine statistically significant differences, represented by different letters. ⁷ Commercial feed, Otohime EP3, 48% protein/10% lipids (Reed Mariculture).

Table 5. Whole-body composition (on wet weight basis) of yellowtail snapper fed a diet containing 36% protein with varying lipid levels for Trial 2. One-way analysis of variance (ANOVA) was used to determine significant differences (*p* < 0.05), represented by different letters. Analysis was performed by MidWest Laboratories, Inc., Omaha, NE, USA.

Dietary Lipids	Moisture %	Protein (Crude) %	Fat %	Ash %
7	69.50 ^a	17.90	7.86 ^b	4.41
10	68.48 ^{a,b}	17.23	9.12 ^{a,b}	4.09
13	66.83 ^b	17.80	9.34 ^{a,b}	4.66
16	66.75 ^b	17.28	10.66 ^a	4.66
PSE ¹	0.37	0.17	0.34	0.11
<i>p</i> -value	0.0049	0.4188	0.012	0.2485
Pro48:Lip10	66.35	18.23	10.70	5.04

¹ PSE = Pooled standard error.

4. Discussion

Understanding and exploring nutritional requirements is crucial in the formulation and optimization of diet development for any species. Given the limited data on yellowtail snapper, this study aimed to establish baseline data for yellowtail snapper nutrition. The range of reference diets previously tested for snapper *Pagrus auratus* in various studies [19–22] makes it difficult to formulate a standard reference diet for snapper to be used in nutritional studies. These diets varied widely, making it challenging to give consistent recommendations on a reference diet. In fact, the spotted rose snapper *Lutjanus guttatus* was reported to have desirable growth performance when fed diets having 470.4 to 529.4 g/kg and 88.8 to 100 g/kg protein and lipids [25]. All of these values are relatively high when compared to the proposed levels of the current study, possibly indicating over formulation for these species as well.

In the present study, no significant differences were observed in most growth parameters across all experimental diets in both trials, except for ANPR in Trial 2. This suggests that all experimental diets provided adequate levels of dietary protein to support growth. This contrasts with other studies which reported that lower protein diets can result in a deficiency of essential nutrients [6,13], which can have an impact on protein synthesis and deposition [26].

ANPR was observed to decrease as protein levels increased, with the highest retention observed in the 7% lipid diet and the lowest in the 16% lipid diet. This is supported by trials conducted in other species, where excessive dietary protein did not increase growth and could even reduce it. This is likely due to a compromise in the fish's immune system and other protective mechanisms, which may be affected by excessive absorption of protein and specific amino acids [27]. These results were observed in cobia (*Rachycentron canadum*) [28], Arctic charr (*Salvelinus alpinus*) [11], and gilthead seabream (*Sparus aurata*) [10]. Excesses of dietary protein not only increase the cost of feeds, but also contributes to environmental pollution so it should be avoided whenever possible.

Although not significantly different, yellowtail snapper fed 40% protein had the highest weight gain % and total dry feed offered per fish (feed intake) as opposed to fish fed 44% protein, despite the fact of it having a higher protein content (Trial 1). This result would suggest that the diet containing 40% protein is the most efficient level for growth, even though no significant differences were observed for all proximate composition parameters, including crude protein, moisture, fat, and ash (Table 3). This is supported by other studies conducted with marine species, such as Atlantic cod *Gadus morhua* [29] and cobia *Rachycentron canadum* [14], where a wide range of protein and lipid diets was used without impacting proximate composition.

Yellowtail snapper fed Pro36:Lip10 in Trial 2 had better growth performance than fish fed Pro44:Lip6, even though this diet had the lowest protein content among experimental diets. This is supported by various studies on marine fish showing nutritional protein-sparing effects of lipids, where balanced energy and protein levels support optimal growth. A study on Australian snapper *Pagrus auratus* proved no difference in growth performance when fed a commercial diet containing 35% and 51% crude protein due to the protein-sparing effect [19,28,30]. Excessively high dietary lipids, however, can cause a reduction in protein and increased lipid levels in whole-body and liver tissues, as seen in other species like cobia *Rachycentron canadum* [31]. Furthermore, highly vacuolized livers were observed in mangrove red snapper *Lutjanus argentimaculatus* [32]. These things should be considered when discerning optimal lipid inclusion levels for formulated snapper diets.

Lower feed acceptance for yellowtail snapper fed Pro44:Lip6 (Trial 1) may be attributed to high soybean meal replacement. This replacement was proven to induce palatability issues [33] in red drum *Sciaenops ocellatus* [2,34], discus *Symphosodon aequifasciata* [35], and Asian seabass *Lates calcarifer* [36,37] (when fed with high or complete soybean meal replacement. High palatability of the commercial feed emphasizes the importance of including attractive feed components during formulation.

In common snook *Centropomus undecimalis*, improved feed intake and protein efficiency ratio was observed in fish fed diets containing high digestive carbohydrates of 20% cornstarch, indicating a clear protein sparing effect of carbohydrates [38]. This was also reported in rainbow trout *Oncorhynchus mykiss* [39] and silver perch *Bidyanus* [40]. Likewise, lower dietary protein levels of 32–36% were sufficient to support good growth in red snapper *Lutjanus campechanus*, where TGC values of snapper fed 44% protein were not significantly different than fish fed 36% [23,41], which correlates with the results of the present study.

The assimilation and use of dietary protein for growth is affected by several factors such as energy and/or lipid levels. Studies focusing on lipid utilization demonstrated reduction in digestible protein to digestible energy ratio (DP:DE) in cobia *Rachycentron canadum* when fed increasing dietary lipids, resulting in tissue lipid deposition [14]. For Trial 2, yellowtail snapper fed Pro36:Lip13 performed best among all experimental diets

(when excluding the commercial diet) and better than fish fed Pro36:Lip16 in spite of it having higher lipid content.

Commercially produced feeds may have different physical and chemical characteristics from laboratory produced feeds, such as water stability and durability, pellet hardness and nutrient bioavailability, which may influence feed intake and feed utilization [22,26]. Commercial floating pellets have been available for more than a decade, but recently, a larger range of sizes and formulations has been marketed [42]. Actual price and amount of commercially produced feeds vary widely depending on the market and the region in the world where it was produced. Another factor that needs to be considered is feed selection, as its quality is directly associated with the seed output and the production cost [42]. Experimental diets for Trial 2 were compared to a commercial marine fish feed as a reference diet due to the unavailability of a customized commercial feed formulated for yellowtail snapper. Typically, feed manufacturers have not been interested in producing specific broodfish diets because of substantially lower demand [43]. In line with this, the commercial feed, Otohime EP3 commercial feed—Pro48:Lip10—was ground following the same methods used for all other experimental diets to have similar physical characteristics.

Significant differences were observed for the total dry feed offered per fish, with the highest values for yellowtail snapper fed commercial feed (Pro48:Lip10). This may be due to the high amount of fishmeal and other marine products that were incorporated in the feed, making it very palatable and likable for the fish. Nonetheless, the same general trend was still observed as when considering the experimental diets (excluding the commercial diet); the best growth performance is observed for yellowtail snapper fed Pro36:Lip13 and the least growth performance is demonstrated in fish fed Pro36:Lip16. This promotes sustainability without compromising growth.

5. Conclusions

Based on growth performance and feed utilization parameters among all protein and lipid levels for Trial 1, the yellowtail snapper was able to effectively utilize practical diets with Pro36:Lip10 and Pro40:Lip10. Interestingly, yellowtail snapper fed the Pro36:Lip10 diet had the highest ANPR% and survival. For Trial 2, commercial feed having Pro48:Lip10 had the best growth performance, feed utilization, and whole-body composition, as expected. When exclusively considering the experimental diets, yellowtail snapper fed Pro36:Lip13 had the best overall performance. The current study aimed to establish baseline data as initial dietary information for yellowtail snapper nutrition and recommends dietary protein within the range of 36–40% and dietary lipid levels of 7–13%, which are lower than protein and lipid levels used currently in commercial diets for marine finfish, suggesting the potential for more sustainable and cost-effective feed formulations. The knowledge gathered from the current study may be helpful for nutritionists seeking to formulate feed based on more sustainable and cheaper feedstuffs and promote sustainable yellowtail snapper aquaculture.

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