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Use of Coontail as a natural phytoremediation feed additive for common carp

by

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Abstract

Ceratophyllum demersum is a submerged, rootless, free-floating macrophyte and natural aquatic phytoremediation. *C. demersum* has high efficiency in absorbing large amounts of nitrogen and phosphorus in the aquatic environment. In this study, the effects of a natural phytoremediation *Ceratophyllum demersum* meal, as a dietary supplement, on growth performance, feed utilization, and body composition of common carp (*Cyprinus carpio* L.,1758) were investigated. This was the first attempt to use coontail as a feed additive for common carp. Four isonitrogenous (38% crude protein), isolipidic (8% crude lipid), and isoenergetic (18 kJ g^{-1}) diets were formulated control group (0%), CM5 group (5%), CM10 group (10%) and CM15 group (15%) in feed. Each dietary treatment was administered to triplicate in groups of 15 fish (~19 g). Carp were hand-fed to apparent saturation three times a day (09.00; 12.00 and 17.00). There was no difference between the control diet and 5% CM in terms of growth (final weight, weight gain, specific growth rate) and feed utilization (feed conversion rate, feed intake, feed efficiency) ($p < 0.05$). Increasing algae levels in diets improved protein levels in body compositions while decreasing lipid. There was no sign of appeared fish health indicators (disease or deformity) in all diet groups. This study results showed that instead of soybean meal, 5.75% and 6.07% as natural phytoremediation, *C. demersum* optimized the best growth and feed utilization performance in carp diets.

Key words: Natural phytoremediation, *Ceratophyllum demersum* meal, common carp, soybean meal, growth and feed performance, body composition

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1. Introduction

In the last 50 years, eutrophication in inland waters has increased drastically around the world (Padedda et al. 2022). Eutrophication, which has become the subject of both academic and public debate, is fed by nutrients in the form of widespread agricultural loading and point sources of industrial and municipal wastewater (Bonsdorff 2021). Eutrophication of aquatic ecosystems results from high concentrations of nutrients, particularly phosphates and nitrates. Coastal water bodies are exposed to pollution from agriculture, industrial facilities, sewage systems, and water treatment plants. Agricultural activities, which are the source of more than 40% of all pollution, gradually upset the balance of water quality (EAA 2015). Fertilizer consumption (N, P, and K) in all countries of the world is predicted to increase from 166 million tons to 263 million in the forty-five-year period from 2005 to 2050, (Alexandratos & Bruinsma 2012). Biologically usable in inland waters can disrupt the balance of the ecosystem, especially phosphorus and nitrogen loads.

Köyceğiz Lake reaches the Mediterranean Sea via the Dalyan Lagoon canal system. Agriculture, tourism, forestry, and fishing are the main sources of economic income in the area of the Lagoon. Fertilization is a common practice in agricultural lands (Gürel 2000). It is calculated that approximately 108 thousand kg of nitrogen and 9.7 thousand kg of phosphorus enter Dalyan Lagoon annually, even if fertilization is undertaken conscientiously and in accordance with good agricultural practices (Ekdal 2008).

Conventional treatment technologies to remove wastewater pollutants from the receiving aquatic environment are both costly and time-consuming. In contrast, phytoremediation is a cost-effective, green emerging technology with long-term viability (Ali et al. 2020).

Aquatic phytoremediation has the potential to be used both as a management strategy for the removal of pollutants from surface waters and for the recovery of nutrients. Macrophytes in particular are used for aquatic phytoremediation. Macrophytes take up N mainly in the form of nitrate (NO_3) and ammonium (NH_4), while P is taken as phosphate (PO_3). *Ceratophyllum* (coontail) is a common genus of rootless submerged plants (Fletcher et al., 2020). *Ceratophyllum demersum* is a good phytoremediator in removing pollutants such as nitrate and phosphate and in the treatment of wastewater (Enas & Dunya 2021). The obtaining of a quantitative reduction of nutrients in domestic wastewater and an increase in net primary productivity value revealed that *C. demersum* is an efficient aquatic plant as phytoremediation (Patel & Kanungo 2010). Foroughi et al. (2013) state that *C. demersum* reduces ammonium and nitrate by more than 62% and 41.66%, respectively, and is one of the best biological methods to treat wastewater with high efficiency in a short time. *C. demersum* has high efficiency in absorbing large amounts of nitrogen (Yaseen & Azawey 2021). *C. demersum* with removal rates of 35% and 73% are potential candidates for total nitrogen and total phosphorus removal, respectively (Dai et al. 2012).

The total world fishery production in 2020 was 178 million tonnes, of which 87.5 million tons was obtained from aquaculture. The aquaculture sector is considered to be the fastest-growing sector among the food production sectors. The common carp is one of the most widely-cultured species of freshwater fish in the world (FAO 2022). World aquaculture production of common carp was close to 4.3 million tonnes in 2020, representing 8.6% of total inland aquaculture production of fish and 4.8% of total aquaculture production. Macro algae (generally known as seaweed) provide a novel and value-added dietary ingredient in fish diets (Wan et al. 2018). The total production of aquatic algae was 35.01 million tonnes (USD 16.5 billion) in 2020 (FAO 2022). Mustafa et al. (1995) stated that a small amount of algae added to the fish diet significantly improves growth, lipid metabolism, body composition, and disease resistance. Also, Chopin & Tacon (2021) revealed that the use of different seaweed raw materials in the diets of fish, crustaceans, mollusks, sea urchins, and sea cucumbers generally positively affects the growth of aquatic organisms at the level of 5–15%. Therefore, the production of algae is being developed. Algae meals, which are used as alternatives in fish diets at different rates, can significantly reduce feed prices when considered economically (Saleh 2020).

The first study on the effect of aquatic weeds on the growth performance of fish was carried out by Venkatesh & Shetty (1978). In their study, *Hydrilla*, *Ceratophyllum* and *Azolla*, a bioremediation aquatic macrophyte, is a potential feed raw material with 22.5% protein and 4.5% lipid (Anitha et al. 2016). *Azolla cristata* protein grown in aquaculture wastewater can replace up to 10% fish meal protein in the diet of Pangasius catfish juveniles without negatively affecting feed intake, growth, survival, and body composition (Rahmah et al. 2022). Goswami et al. (2022) found that the inclusion of *Lemna minor* in the diet of carp enhanced the nutritional value of the carp by increasing protein, lipid, amino acids and n-3 PUFA contents in *Cyprinus carpio* fingerlings. Irabor et al. (2022) found that a 40% inclusion level of *Lemna* *minor* meal in the diet for 56 days is best for optimum growth performance for juvenile *C. gariepinus*, with any negative effect. In another study, Fierdelmundo et al. (2022) found that duckweed meal derived from *Lemna minor* can be included in the feed for the rainbow trout without negative effects on the growth performances at 20% of the protein substitution.

C. demersum (Ceratophyllaceae), which is considered native to many parts of the world, including Türkiye, is a submerged, rootless and free-floating macrophyte. It has a high vegetative spreading capacity (Dogan & Demirors 2018). *C. demersum* is called "tilki kuyruğu" in Türkiye. *C. demersum* is found in the Dalyan Lagoon canal system as natural phytoremediation.

This study aims to demonstrate the effects of *C. demersum* as phytoremediation on the growth performance, feed evaluation, and body composition of common carp (*Cyprinus carpio*) fingerlings and to fill the lack of information about the result of feeding carp with *C. demersum*.

2. Materials and methods

2.1. Preparation of Experiment Feeds

C. demersum, phytoremediation used in this study, was freshly collected from the Köyceğiz – Dalyan Lagoon. Collected *C. demersum* samples were dried on blotting papers spread out on the tables in the laboratory at an average room temperature of 25°C for 5 days. The dried macroalgae samples were ground into a meal in a gristmill located in the Muğla-Ortaca industrial zone.

Four isonitrogenous (38% crude protein), isolipidic (8% crude lipid), and isoenergetic (18 kJ g^{-1}) diets were formulated to meet the nutritional needs of common carp. The feed experiment included a control group with no *C. demersum* meal (C), and 5%, 10%, and 15% *C. demersum* meal instead of soybean meal groups (CM5, CM10, and CM15). Fish meal, soybean meal, wheat meal, fish oil, *C. demersum* meal, wheat gluten, vitamin-mineral premixes, and antioxidants were used in experimental diets. The composition of the *C. demersum* meal, fish meal, soybean meal, wheat meal, and wheat gluten with an essential amino acid profile is given in Table 1. The nutritional and chemical compositions of the diets are reported in Table 2.

Dry ingredients were mixed according to weight and homogenized for 16 minutes. Fish oil and water were added to the mixture which was then pulped and converted to a 2mm diameter pellet after being passed through a pelletizer. The pellets were prepared using a constant temperature (about 70°C) feed-making machine. Prepared feeds were crumbled into a dried material and maintained at 4°C for three days until use.

2.2. Experiment Planning

Common carp (*Cyprinus carpio*) fingerlings were obtained from the Institute of Mediterranean Fisheries Research Production and Training (Antalya-Kepez/Türkiye). The fish that were provided in the transfer was placed in a 500 l fiberglass tank at the research unit of Muğla Sıtkı Koçman University Ortaca Vocational School in Muğla, Türkiye. 15 days before the test, the fish were acclimated in the experiment tank and fed 3 times per day (a commercial diet, crude protein 38%, crude lipid 8%). 180 fish were randomly distributed into 225 l fiberglass tanks with 15 fish each (mean weight 18.89 ± 0.14 g SD). Each tank had an open recirculation system. The fingerlings were fed four experimental diets lasting 52 days. During the

Table 1

Proximate composition and essential amino acid profile of feed ingredients in test diets

Proximate Analysis	CDM*	SBM	FM	WM	WGM
Dry matter (% in an original matter)	89.57	89	92	88	89
Crude Protein	15.78	44	65.4	11.7	80.7
Crude Lipid	1.80	1.5	7.6	1.2	1.5
Crude Fibre	18.61	7.3	1.0	1.3	0.5
Ash	18.96	6.3	14.3	0.4	0.7
NFE[1]	34.42	29.90	3.70	73.40	5.60
Gross Energy (kJ g^{-1})[2]	10.22	15.83	18.58	15.74	20.10
Calcium	0.22	0.03	0.63	0.03	0.14
Phosphorus	0.38	0.07	0.39	0.34	0.26
EAA (DM %)[3,4]					
Arginine	0.45	3.23	3.68	0.86	3.80
Histidine	0.04	1.17	1.56	0.39	2.00
Isoleucine	0.70	1.99	3.06	0.51	3.70
Leucine	1.09	3.42	5.00	0.92	6.30
Lysine	0.95	2.83	5.11	0.58	4.90
Methionine	0.18	0.61	1.95	0.19	1.60
Phenylalanine	0.64	2.18	2.66	0.55	4.50
Threonine	0.70	1.73	2.82	0.46	1.60
Tryptophan	—	0.61	0.76	0.25	1.05
Valine	0.76	2.40	3.51	0.69	4.00

CDM - *C. demersum* meal, SBM - Soybean meal, FM - Fish Meal, WM - Wheat Meal, WGM - Wheat Gluten Meal

[1] Nitrogen-free extracts (NFE) = Dry matter % – (lipid (%) + protein (%) + fiber (%) + ash (%)

[2] Calculated using the factors: carbohydrates, 4.1 kcal g^{-1}; protein, 5.5 kcal g^{-1}; lipid, 9.1 kcal g^{-1} (New 1987), and use a factor of 4.184 to convert to kJ.

[3] Essential amino acids values of soybean meal, fish meal, and wheat meal were obtained from NRC (2011).

[4] Essential amino acids values of *C. demersum* meal were obtained from Laining et al. (2016).

* CDM crude protein, crude lipid, crude fibre, ash, and dry matter values analysed in our study; SBM, FM, and WM values obtained from NRC (2011).

experiment, the fish were fed 3 times a day (8:00 a.m., 12:00 noon, and 4:30 p.m.) to satiation. Uneaten feed and feces were siphoned twice daily.

Dissolved oxygen, temperature and pH in the water system were measured daily using the Hach-Lange HQ40 D model multiparameter device. The N-NO_2- and N-NO_3 content of water were measured weekly using Noratex test kits. The mean values were as follows: mean water temperature 20.5°C ± 2, 8.68 mg l^{-1} ± 1 dissolved oxygen, 8.3 ± 0.2 pH, 0.1 ppm N-$NO_{2,}$ and 0.5 ppm N-NO_3.

Table 2

Ingredients and proximate composition of the four experimental diets

<table>
<tr><th rowspan="2">Ingredients (%)</th><th colspan="5">Diets</th></tr>
<tr><th colspan="2">Control</th><th>CM5</th><th>CM10</th><th>CM15</th></tr>
<tr><td>Fish Meal</td><td colspan="2">300</td><td>305</td><td>305</td><td>305</td></tr>
<tr><td>Soybean Meal</td><td colspan="2">360</td><td>342</td><td>324</td><td>306</td></tr>
<tr><td>Wheat Meal</td><td colspan="2">205</td><td>200</td><td>200</td><td>190</td></tr>
<tr><td>Fish Oil</td><td colspan="2">100</td><td>100</td><td>100</td><td>100</td></tr>
<tr><td><i>C. demersum</i></td><td colspan="2">0</td><td>18</td><td>36</td><td>54</td></tr>
<tr><td>Wheat Gluten</td><td colspan="2">20</td><td>20</td><td>20</td><td>30</td></tr>
<tr><td>Vit-Min Mix</td><td colspan="2">10</td><td>10</td><td>10</td><td>10</td></tr>
<tr><td>Antioxidant</td><td colspan="2">5</td><td>5</td><td>5</td><td>5</td></tr>
<tr><th colspan="6">Proximate analysis (% DM)</th></tr>
<tr><td>Dry matter</td><td colspan="2">90.55</td><td>90.92</td><td>90.91</td><td>91.02</td></tr>
<tr><td>Crude protein</td><td colspan="2">37.99</td><td>37.90</td><td>37.54</td><td>37.87</td></tr>
<tr><td>Crude lipid</td><td colspan="2">8.00</td><td>8.06</td><td>8.06</td><td>8.06</td></tr>
<tr><td>Crude Fibre</td><td colspan="2">1.19</td><td>1.94</td><td>2.63</td><td>3.79</td></tr>
<tr><td>Ash</td><td colspan="2">5.71</td><td>6.64</td><td>7.76</td><td>8.40</td></tr>
<tr><td>NFE[1]</td><td colspan="2">36.66</td><td>36.38</td><td>34.92</td><td>32.90</td></tr>
<tr><td>Calcium</td><td colspan="2">0.33</td><td>0.34</td><td>0.35</td><td>0.36</td></tr>
<tr><td>Phosphorus</td><td colspan="2">0.3</td><td>0.3</td><td>0.28</td><td>0.26</td></tr>
<tr><td>Gross Energy (kJ g^{-1})[2]</td><td colspan="2">18.46</td><td>18.03</td><td>17.70</td><td>17.43</td></tr>
<tr><th colspan="6">EAA (g 100 g^{-1} DM)</th></tr>
<tr><td>Amino Acids</td><td>Carp[3]</td><td>Control</td><td>CM5</td><td>CM10</td><td>CM15</td></tr>
<tr><td>Arginine</td><td>1.7</td><td>2.52</td><td>2.48</td><td>2.43</td><td>2.41</td></tr>
<tr><td>Histidine</td><td>0.8</td><td>1.01</td><td>0.99</td><td>0.97</td><td>0.97</td></tr>
<tr><td>Isoleucine</td><td>1.0</td><td>1.81</td><td>1.80</td><td>1.78</td><td>1.79</td></tr>
<tr><td>Leucine</td><td>1.5</td><td>3,05</td><td>3,02</td><td>2,98</td><td>2,99</td></tr>
<tr><td>Lysine</td><td>2.2</td><td>2.77</td><td>2.76</td><td>2.72</td><td>2.73</td></tr>
<tr><td>Methionine</td><td>0.3</td><td>0.88</td><td>0.88</td><td>0.87</td><td>0.88</td></tr>
<tr><td>Phenylalanine</td><td>1.3</td><td>1.79</td><td>1.77</td><td>1.74</td><td>1.75</td></tr>
<tr><td>Threonine</td><td>1.5</td><td>1.60</td><td>1.59</td><td>1.57</td><td>1.56</td></tr>
<tr><td>Tryptophan</td><td>0.3</td><td>0.52</td><td>0.51</td><td>0.50</td><td>0.50</td></tr>
<tr><td>Valine</td><td>1.4</td><td>2.14</td><td>2.12</td><td>2.09</td><td>2.10</td></tr>
</table>

[1] Nitrogen-free extracts (NFE) = Dry matter % – (lipid (%) + protein (%) + fibre (%) + ash (%)

[2] Calculated using the factors: carbohydrates, 4.1 kcal g^{-1}; protein, 5.5 kcal g^{-1}; lipid, 9.1 kcal g^{-1} (New 1987), and use a factor of 4.184 to convert to kJ.

[3] Requirements according to NRC (2011).

2.3. Sampling and method of analysis

All experimental feeds and fish samples were analyzed for the chemical composition according to the Association of Official Analytical Chemistry (AOAC 2002; AOAC 2002b). Dry mass was determined by oven drying at 105°C for 24 hours to constant weight; ash content was determined by firing in a muffle furnace at 550°C for 4 hours; Crude protein was determined by Kjeldahl protein unit. Crude lipid was determined by the ether extraction method. Before starting the experiment, 5 fish from the first batch were chosen to be subjected to hypothermia in the refrigerator, stored in polyethylene bags, and frozen (-20°C) for subsequent body composition analysis. At the end of the feeding trial, 5 fish were randomly removed from each tank (15 fish per treatment), killed as described above, and stored for analysis. Samples were prepared by homogenizing whole fish in a blender prior to analysis. All analyses were performed in triplicate.

2.4. Calculation of growth parameters

Growth parameters and feed utilization protein efficiency were calculated with the following equations:

$$\text{Weight Gain (\%)} = \frac{\text{Final wet weight (g)} - \text{Initial wet weight (g)}}{\text{Initial wet weight (g)}} \times 100$$

$$\text{Specific Growth Rate (\% Day}^{-1}\text{)} = \frac{\text{Ln final average weight (g)} - \text{Ln initial average weight (g)}}{\text{trial days}} \times 100$$

$$\text{Feed Conversion Rate} = \frac{\text{Total˝Feed Intake (g)}}{\text{Weight Gain (g)}}$$

$$\text{Daily feed intake (g fish}^{-1}\text{ day}^{-1}\text{)} = \frac{\dfrac{\text{Total amount of feed per tank}}{\text{Number of fish in the tank}}}{\text{Total trial days}}$$ (Ponzoni et al. 2013),

$$\text{Feed Efficiency Value} = \frac{\text{Weight Gain (g)}}{\text{Total Feed intake (g)}}$$

$$\text{Protein Efficiency Rate} = \frac{\text{Weight Gain (g)}}{\text{Protein Intake (g)}}$$

$$\text{Survival Rate (\%)} = \frac{\text{Number of fish harvested}}{\text{Number of fish stocked}} \times 100$$ (Biswas et al. 2011),

2.5. Ethical Approval

Ethics statement all animal care, welfare, and handling procedures in the present study protocol were approved (Protocol Number 64583101/127) by the Animal Experiments Local Ethics Committee of Aydın Adnan Menderes University of Türkiye. This manuscript was produced from *Ümmühan* Kızıloğlu's master thesis.

2.6. Statistical Analyses

Statistical differences among the experimental groups were tested by one-way ANOVA followed by post hoc Tukey's test ($p < 0.05$) by using SPSS for Windows 22.0" program. The multiple regression for specific growth rate and feed conversion rate between dietary *C. demersum* levels has been applied in R open-source statistics software.

3. Results

3.1. Growth performance and survival

The results of growth rates and survival percentages of fish fed on four different feeds (Basal diet, three coontail diets) are shown in Table 3. There was no statistical difference ($p > 0.05$) between the control and CM 5 in terms of final weights. However, CM10 and CM15 were observed to be poorer in terms of both fish final weight and weight gain. The specific growth rate values supported this trend. SGR was the same in the control and 5% CM, while 10–15% CM was lower. There is no statistically significant difference between the survival rates of the groups ($p > 0.05$).

3.2. Feed consumption and feed utilization

The amount of feed consumption between the experimental groups was not significant ($p > 0.05$). Mean daily feed intake ranged between 0.45 and 0.47 g/fish/day. The best feed conversion ratio (FCR) values differed significantly ($p < 0.05$) between the control group and 5% CM (1.66-1.70) when compared to fish fed on 10% CM and 15% CM (2.38–2.75) (Table 3).

3.3. Biochemical composition

The results of the dietary effect on the body chemistry of the *C. carpio* composition at the conclusion of the feeding trial are presented in Table 4. A significant difference between body crude protein

Table 3

Growth performance and feed utilization parameters of Common Carp fed experimental diets

Parameters	Units	Control	CM5	CM10	CM15
Initial Weight	g	19.08 ± 0.13^{a}	18.88 ± 0.04^{a}	18.87 ± 0.06^{a}	18.73 ± 0.15^{a}
Final Weight		33.83 ± 0.07^{a}	33.42 ± 0.38^{a}	29.43 ± 1.70^{b}	27.30 ± 0.55^{b}
Weight Gain		14.75 ± 0.12^{a}	14.54 ± 0.42^{a}	10.56 ± 1.65^{b}	8.56 ± 0.64^{b}
Weight Gain	%	77.31 ± 1.03^{a}	77.01 ± 2.38^{a}	55.96 ± 8.62^{b}	45.76 ± 3.69^{b}
SGR		1.1 ± 0.00^{a}	1.09 ± 0.02^{a}	0.86 ± 0.09^{b}	0.71 ± 0.04^{b}
Daily feed intake	g fish^{-1} day^{-1}	0.472 ± 0.006^{a}	0.476 ± 0.007^{a}	0.475 ± 0.005^{a}	0.450 ± 0.004^{b}
FCR		1.66 ± 0.03^{b}	1.70 ± 0.05^{b}	2.38 ± 0.25^{a}	2.75 ± 0.23^{a}
Feed Efficiency Value		0.60 ± 0.01^{b}	0.58 ± 0.02^{b}	0.44 ± 0.04^{a}	0.36 ± 0.03^{a}
Protein efficiency rate		1.75 ± 0.45	1.69 ± 0.61	1.22 ± 0.23	1.00 ± 0.35
Survival Rate	%	97.77 ± 3.14^{a}	95.55 ± 3.14^{a}	95.55 ± 4.44^{a}	97.77 ± 3.14^{a}

Values refer to mean ± standard deviation. Values expressed in different exponential letters in the same row are statistically different from each other ($p < 0.05$).

Table 4

Carcass composition of common carp fed diets containing different levels of *C. demersum*

Parameters	Units	Control	CM5	CM10	CM15
Crude Protein	%	16.55 ± 0.32^{b}	17.46 ± 0.25^{a}	17.21 ± 0.51^{a}	16.76 ± 0.25^{b}
Crude Lipid		9.68 ± 0.62^{a}	8.46 ± 0.55^{a}	6.59 ± 0.76^{b}	6.75 ± 0.45^{b}
Moisture		71.86 ± 3.21	71.97 ± 1.55	74.19 ± 1.11	74.57 ± 2.25
Crude Ash		1.91 ± 0.11	2.11 ± 0.20	2.01 ± 0.10	1.98 ± 0.9

Values refer to mean ± standard deviation. Values expressed in different exponential letters in the same row are statistically different from each other ($p < 0.05$).

of the 5% CM and 10% CM treatments and control and 15% CM was observed ($p < 0.05$). A slight decrease in body crude lipid content was noted in fish-fed diets supplemented with coontail compared to the control group. Nevertheless, there was no statistical difference between the control and 5% CM. Body moisture and ash composition were similar between the groups ($p > 0.05$).

4. Discussion

The present study reports the first use of a natural phytoremediation *C. demersum* in common carp diets. We primarily investigated the efficiency of the substitution of soybean meal with *C. demersum* meal in common carp feed and the resulting effects on the growth, feed utilization performance, and biochemical composition of the fish fed on the substitute feed. As a result of the substitution with *C. demersum* meal, more than 5% of growth was suppressed. This was about 21% and 31% less growth for 10% *C. demersum* and 15% *C. demersum* than for the control and 5% *C. demersum*. In this study, fish growth performance and feed utilization tended to decrease with the increase of algae rates in diets. The regression-specific growth rate and feed conversion rate indicated that the most suitable soybean meal replacement level by *C. demersum* meal was 5.75% and 6.07% respectively for the best growth of common carp juveniles (Figure 1 and 2).

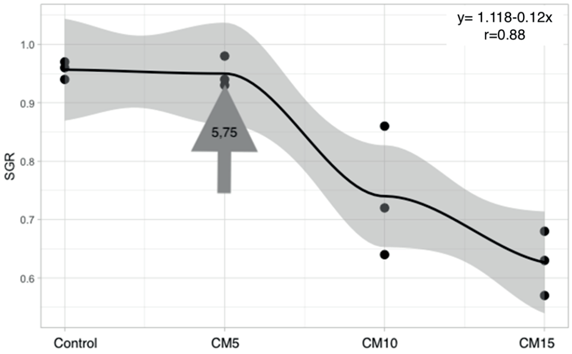

Figure 1

Specific Growth Rate (%)

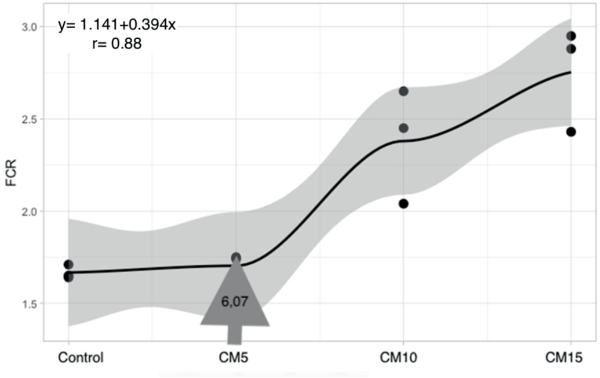

Figure 2

Feed Conversion Rate

The findings of Balkhasher et al (2021) showed that the use of *C. demersum* in diets of Nile tilapia was relatively better than *P. amplifolius*. However, the substitution of 33% *C. demersum* instead of wheat bran meal reduced the final weight by 10.6% and PER by 42.3% compared to the control group. Goswami et al. (2022) aimed to determine the optimum dietary *Lemna minor* meal inclusion level in *C. carpio* diets. They reported that diets supplemented with duckweed had a positive effect on growth. Fierdelmundo et al. (2022) showed that no adverse effects were observed in the mean body weight and weight gain of rainbow trout when the fish were fed diets containing up to 20% duckweed meal. Irabor et al. (2022) noted that the inclusion of 40% duckweed meal in the diet of juvenile catfish is best for optimum growth performance without any adverse effects. In another aspect, Komara et al (2022) showed that coontail (*C. demersum*) is an inexpensive, safe, widely available and environmentally friendly carbohydrate source for Nile tilapia culture in the biofloc system. In another investigation, Catfish was found to have better growth performance when fed with 'Azolla' grown from wastewater, replacing up to 10 g kg^{-1} of fishmeal protein (Rahmah el.al 2022). In contrast, Laining et al. (2016) reported rates parallelling those of the control group when using up to 30% of *Ceratophyllum* weed meal in the growth diets of rabbitfish. The amino acid content of the feed ingredients can be considered among the reasons for these growth differences. When the amino acid profile of raw materials and diets is examined (Table 1), *C. demersum* shows possible deficiencies in all EAAs. Therefore, *C. demersum* seems to be a source of protein with lower nutritional status than SBM. In addition, more than 5% of diets can cause poor growth and feed utilization in fish. Reasons for this include EAA deficiencies or imbalances in the amino acid profile. Another possibility is the availability of anti-nutrients in the algae content (Azaza et al. 2008; Yildirim et al. 2009; Natify et al. 2015).

Anti-nutrient factors can bind to nutrients and affect their usefulness. Examples of these substances are saponins, tannins, phytic acid, gossypol, lectins, protease inhibitors, and amylase inhibitor (Samtiya et al. 2020). Although most of the anti-nutrients in fish diets do not cause death, they may adversely affect biomass (Francis et. 2001). Norambuena et al. (2015) also stated that complex polysaccharides in algal products can adversely affect the digestibility of nutrients. Omnes et al. (2017) demonstrated that tannin supplementation of upwards of 10 g kg^{-1} in European sea bass diets reduces protein digestion, while growth performance above 20 g kg^{-1} significantly decreases. Although there are anti-nutrient substances such as lectin in marine algae, they are less common than in terrestrial plants (Rogers & Hori 1993). The main point here is that knowing the threshold level of these components can eliminate their harmful effects.

The whole-body composition of fish is often used as an indicator of fish health and meat quality (Ahmed 2018). In this study, the higher protein level results in increased body composition of fish due to the increase of aquatic macrophyte in the diets, whereas the rise in the *C. demersum* level reduced the lipid. In contrast, there was not only an increase in protein in common carp, but lipid contents also showed linear relationships ($p < 0.05$) with increasing the amount of duckweed included in the diet (Goswami et al. 2022). Similarly, Rahmah et al. (2022) revealed that the body composition of Pangasius catfish protein and lipid levels increased when using 10% *Azolla cristata* level in catfish diets. Fierdelmundo et al. (2022) found that fish fed with 20% *Lemna minor* meal tended to show lower protein and higher lipid content than the control and 10% group. On the other hand, Irabor et al (2022) determined that with 40% and higher levels of duckweed in the diets, both the protein and fat levels decreased in catfish biochemistry. Mobilization of these lipid reserves is an important parameter for fish (Ergun et al. 2009). The existence of an effect of vitamin C on lipid metabolism has been emphasized (Ji et al. 2003). Algae constitute a high concentration of vitamins and minerals. For example, the concentration of vitamin C may vary from 38 µg g^{-1} (*Ulva* sp.) to 362 µg g^{-1} (*Sargassum* sp.) dry weight (Garcı´a-Casal 2007). The ascorbic acid values of some algae are well documented (Kovacık 2019). Kovacik et al. (2017) found 2669 µg g^{-1} (dry weight) amounts of ascorbic acid in *C. demersum*. The abundance and fluctuation of ascorbic acid in algae draw attention in terms of non-traditional sources of Vitamin C (Garcia-Casal 2007).

Throughout the experimental study, we did not detect a difference in longevity in all diet groups and there was no visual sign of disease or deformity. This research was the first attempt to supplement the diet of carp fish with the natural phytoremediation of *C. demersum*. Up to 5% of this aquatic macrophyte substance can be substituted for soybean meal in *Cyprinus carpio* diets without any adverse effects. Further investigation of the different incorporation rates of *C. demersum*, which is one of the macrophytes, in the diets of other species besides carp will be of interest in the elimination of some of the gaps in the research.

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