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Proximate, amino acid, fatty acid and mineral composition of Grey mullet, Mugil cephalus L: A comparative study between the culture and wild resources in different size groups and potential contribution to nutritional security

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Abstract

Mugil cephalus is widely distributed across all oceans and it is a potential candidate species for aquaculture. Nutrient profiling in terms of proximate, amino acid, fatty acid and mineral composition was analyzed in muscle tissue of *M. cephalus* sourced from culture and wild with four different sizes (100–150 g, 151–250 g, 251–500 g and > 500 g). Results of proximate composition revealed that both the factors (resource and size) had a significant (P < 0.05) effect on moisture, crude lipid, total ash and gross energy but not on crude protein and crude fibre. Though the amino acid composition was not influenced by resource and size, the level of essential amino acids of ideal protein was higher than the recommended level by FAO/WHO for phenylalanine (43.29–45.60%), tryptophan (36.60-39.02%) and lysine (33.61–41.51%). Fatty acids like C14:0, C16:0, C18:0, C18:0, C18:2c and γC18:3 was significantly (P < 0.05) high in cultured species compared to wild caught fish, irrespective of the size groups and the reverse was true for αC18:3, C20:4 and C20:5. The amount of calcium, phosphorus, sodium and potassium was significantly (P < 0.05) higher by 10.68, 5.82, 9.31 and 6.93% in wild caught fish than its counterpart. The potential contribution of this fish to nutritional security in terms of its daily value (DV) was calculated for one serving of 100 g fish to adult human being. Results revealed that lysine, methionine, threonine, tryptophan and EPA + DHA were considered as outstanding nutrients in this fish, irrespective of their source and size as their DV crossed > 70%. Similarly, isoleucine, leucine, phenylalanine and valine could also able to meet 50–70% of the daily needs of adult human indicating the nutritional richness of both wild and cultured *M. cephalus* regardless of its size.

Introduction

Fish is a relatively cheaper food material with higher quality than the meat sources, which plays a crucial role in global Food Nutrition Security (FNS), especially in low and middle income countries¹. Fish serves a healthy, safe, nutritious and balanced diet to human, as it is rich in valuable nutrients (protein with essential amino acids (EAA), long chain n-3 polyunsaturated fatty acids (LC n-3 PUFA), minerals mainly micro elements). Therefore, its consumption plays an important role in reducing global burden of disease. Alasalvar et al² stated that LC n-3 PUFA cannot be synthesized in human and should be supplemented by diet. As this is rich in fish, helps in neural development in both *utero* and first few years after birth in infants³. In addition, consuming PUFA plays a vital role in controlling various diseases and disorders like aggression, arrhythmias, cancer, coronary heart failure, depression, hypertension, inflammation, psoriasis, etc⁴. WHO⁵ and FAO/IFAD/WFP⁶ reported that around 870 million peoples are affected by protein-energy malnutrition globally and it is nearly 80% in developing countries alone. As fish is an important protein source, its prolonged use can reduce a lethal form of malnutrition⁷. Likewise, certain functional amino acids involved in the process of wound healing and act as an anti-mutagenicity and anti-oxidant⁸. Similarly, element content of fish would be most beneficial to human, as they play vital roles in various metabolic pathways⁹.

The capture fisheries production has become static (96.4 million tonnes in 2018), which could not meet the demand for fish due to the geometric growth of global population¹⁰. However, the expansion of aquaculture production (114.5 million tonnes in 2018) could able to bridge the gap between the demand and supply worldwide. FAO10 reported that the aquaculture sector grown by 527% in 2018 compared to 1990, which alone contributes by 54.46% of the total global fish production with currently valued at USD 263.6 billion¹¹. As in the world, Indian aquaculture sector has also been evolved with considerable diversification in terms of species. Though the Indian major carps have dominated, certain other fishes have also been cultured in India traditionally. One among them is Grey mullet (Mugil cephalus L), and it is predominant in the state of West Bengal, Andhra Pradesh, Tamil Nadu and Kerala in India. M. cephalus has a very wide distribution in all the oceans, estuaries and brackishwater of tropical and sub-tropical regions from 42° N to 42° S. Their catches have increased as a result of a growing market in the south-eastern United States, Gulf coast, Asian and Mediterranean markets with good nutrient profiles 12. Jannathulla et al 13 reported that the nutrient composition of aquatic animals greatly varies, not only between the species and even within the groups, which is mainly due to the variation in feed and biological resource present in the waterbody. In addition, various factors would also influence the nutrient profiling of fish, the most important are age and sources of collection 14. In this context, various methods have been developed by Nutrition Labelling and Education Act (NLEA) in the United States and Association of Analytical Chemist (AOAC) for documenting fundamental knowledge on the nutritional quality of aquatic species. Several studies have already been reported the nutrient compositions of various fin-fishes collected from wild and culture^{14–18}. However, the available data on *M. cephalus* is scarce yet to date. Hence, the present study is aimed to investigate the nutrient composition of muscle tissue in M. cephalus collected from two different sources with four varied size groups. This baseline data would help to facilitate the consumers to realize about the nutritional quality of this fish, by which they could assure about their health, immunity and fitness against the lifestyle disease, which is of great significance to increase the market value of this fish in India and around the globe in the future.

Results

Proximate composition of M. cephalus sourced from both culture and wild with different size groups (Table 1) revealed that both the factors (resource and size) had a significant (P < 0.05) effect on moisture, crude lipid, total ash and gross energy but not on crude protein and crude fibre level. The moisture content was significantly (P < 0.05) high in wild caught fish (74.23%) compared to culture one (72.32%) and was gradually decreased from 75.08–70.57% with increasing size. Wild caught fish had 2.54% of crude lipid and was more than doubled in cultured species (5.44%). Though the lipid content was almost similar in the groups categorized under 100-150 g and 151-250 g (2.44–2.75%), its level was higher by 36% and 141% in 251-500 g and > 500 g groups, respectively. Total ash content found to be high (P < 0.05) in wild caught fish compared to the cultured one, whereas, its level was not affected in the groups had a weight beyond 151 g. Gross energy was significantly (P < 0.05) high in cultured fish (133.03 Kcal/100 g) than wild fish (110.12%) and was gradually (P < 0.05) increased from 107.46-145.27 Kcal/100 g with increasing size.

Table 1
Proximate composition (% wet weight basis) of different size group of *M. cephalus* collected from culture and wild resources.

Particulars	Proximate	Proximate composition									
	Moisture	Crude protein	Crude fat	Crude fibre	NFE	Total ash	(Kcal/100 g)				

Resource (A)							
Culture	72.32 ^b	20.40 ^a	5.44 ^a	0.11 ^a	0.48 ^b	1.20 ^b	133.03 ^a
Wild	74.23 ^a	20.67 ^a	2.54 ^b	0.12 ^a	1.00 ^a	1.42 ^a	110.12 ^b
Size (B)							
100-150 g	75.08 ^a	20.39 ^a	2.44 ^c	0.11 ^a	0.86 ^a	1.17 ^b	107.46 ^c
151-250 g	74.17 ^{ab}	20.94 ^a	2.75 ^c	0.13 ^a	0.65 ^a	1.31ª	111.71°
251-500 g	73.37 ^b	20.45 ^a	4.14 ^b	0.11 ^a	0.56 ^a	1.34ª	121.85 ^b
> 500 g	70.57 ^c	20.36 ^a	6.63ª	0.11 ^a	0.90 ^a	1.42 ^a	145.27 ^a
P-value							
А	< 0.001	0.203	< 0.001	0.373	0.038	< 0.001	< 0.001
В	< 0.001	0.167	< 0.001	0.500	0.690	0.005	< 0.001
AxB	0.011	0.549	< 0.001	0.847	0.700	0.437	< 0.001
SEM (±)	0.269	0.133	0.097	0.001	0.179	0.006	9.337
CV (%)	0.931	2.335	10.255	20.535	74.479	7.629	3.308

Among the essential amino acids (EAA), leucine found to be high followed by lysine and arginine, while the least values were noticed with histidine and tryptophan, whereas in non-essential amino acids (NAA), the highest values were observed for glutamic acid followed by aspartic acid (Table 2). However, no significant differences were observed not only between the resources and also among the size groups. Ideal protein (IP) level, calculated based on the FAO/WHO/UNU¹⁹ values, showed that though there was no much of difference between culture and wild fish, the IP level was increased for all the tested amino acids than the FAO/WHO/UNU¹⁹ recommended values except valine (Fig. 1). The IP level given by FAO/WHO/UNU¹⁹ for Phe + Tyr, lysine and tryptophan are 60, 55 and 10 mg/g, respectively, and was increased by 47.81, 35.73 and 39.17% in cultured fish and by 41.41, 39.50 and 36.33% in wild caught fish. However, the increase was ranged between 11 and 15% for isoleucine, leucine and threonine. A similar trend was noticed in the fish categorized under different size groups. Though the IP level of valine was decreased around by 10%, irrespective of the resource and size, the computed EAAI, based on FAO/WHO/UNU¹⁹ values, did not have any influence on them and was in the range of 0.9251-0.9719 (Fig. 2). Regression performed between culture and wild caught fish, irrespective of the size groups was positively correlated and the equation was $y = 0.005 \times 2-0.0042x + 0.9536$, $R^2 = 0.184$ for cultured fish and $y = 0.002 \times 2-0.0049x + 0.9564$, $R^2 = 0.2696$ for wild caught fish (Fig. 2).

Table 2
Amino acid composition (% wet weight basis) of different size group of *M. cephalus* collected from culture and wild resources.

Particulars	Essenti	al amino	acids													
	Arg	His	lle	Leu	Lys	Met	Phe	Thr	Trp	Val	Ala	Asp	Cyt	Glu	Gly	Pı
Resource (A)																
Culture	1.38	0.26	0.93	1.60	1.52	0.76	1.09	0.93	0.28	0.92	1.30	2.06	0.05	3.14	0.95	0.
Wild	1.42	0.26	0.93	1.61	1.59	0.81	1.04	0.92	0.28	0.93	1.31	2.05	0.05	3.23	0.92	0.
Size (B)																
100- 150 g	1.35	0.26	0.90	1.57	1.50	0.74	1.06	0.91	0.28	0.91	1.28	2.00	0.05	3.16	0.89	0.
151- 250 g	1.44	0.28	0.96	1.64	1.58	0.79	1.10	0.94	0.29	0.93	1.34	2.10	0.05	3.21	0.98	0.
251- 500 g	1.41	0.25	0.93	1.64	1.55	0.80	1.05	0.93	0.28	0.94	1.29	2.07	0.05	3.19	0.93	0.
> 500 g	1.40	0.27	0.92	1.58	1.59	0.81	1.04	0.93	0.28	0.91	1.31	2.06	0.05	3.17	0.94	0.
P-value																
А	0.127	0.936	0.996	0.766	0.076	0.329	0.355	0.749	0.830	0.452	0.701	0.795	0.184	0.184	0.482	0.
В	0.059	0.335	0.387	0.480	0.260	0.721	0.847	0.930	0.969	0.551	0.332	0.358	0.810	0.943	0.232	0.
AxB	0.665	0.353	0.769	0.746	0.827	0.799	0.961	0.858	0.849	0.477	0.945	0.892	0.122	0.643	0.678	0.
SEM (±)	0.001	0.001	0.002	0.006	0.004	0.008	0.011	0.004	0.001	0.001	0.001	0.005	0.001	0.014	0.003	0.
CV (%)	3.635	8.295	5.864	6.146	5.137	15.403	12.790	8.865	8.660	4.857	3.808	4.469	10.003	4.830	8.277	12

Both resources and size have highly influenced (P < 0.05) the tissue fatty acid composition as depicted in Table 3. Among the fatty acids presented, C16:0 was abundant and accounted about 35–40% of total fatty acids, while the least was noticed with both γ and α C18:3. Fatty acids like C14:0, C16:0, C18:, C18:1c, C18:2c and γ C18:3 was significantly (P < 0.05) high in cultured species compared to wild caught fish and the reverse was true for α C18:3, C20:4 and C20:5, whereas there was no significant difference for C22:6 between culture (120.07 mg/100 g) and wild (118.45 mg/100 g) collected fish. The size of the fish had a direct proportion to the muscle fatty acid composition, including eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA). However, the ratios of n-6/n-3 and DHA/EPA were found to be high in 151–250 g and 251–500 g groups, respectively. Fatty acids like C14:0, γ C18:3, C20:4 and C20:5 were significantly (P < 0.05) increased with increasing fish size, whereas there was no significant difference for C16:0, C18:0, C16:1, C18:2c and C22:6 between the fish categorized into 100–150 g and 151–250 g groups. Similarly, no significant difference was observed for C18:1c and C18:2c between the groups had a body weight of 151–250 g and 251–500 g. Among the presented fatty acids, the difference was very conspicuous for C16:0 and was increased by 1103.61 mg/100 g in cultured fish compared to the wild one. Among the size groups, the increase was 1187.39, 1105.80, 730.23 mg/100 g in fish with a body weight of > 500 g compared to the groups of 100–150 g, 151–250 g and 251–500 g, respectively.

Table 3

Major fatty acid composition (mg/100 g wet weight basis) of different size group of M. cephalus collected from culture and wild resources.

Particulars	Major fatt	Major fatty acids												
	C14:0	C16:0	C18:0	C16:1	C18:1c	C18:2c	γC18:3	αC18:3	C20:4	C20:5	C22:6			
Resource (A)														
Culture	124.76 ^a	1585.87ª	453.46ª	610.76ª	728.27 ^a	455.04ª	34.72 ^a	18.80 ^b	60.44 ^b	122.31 ^b	120.07 ^a			
Wild	71.53 ^b	482.26 ^b	241.65 ^b	296.28 ^b	111.09 ^b	44.55 ^b	23.30 ^b	76.53 ^a	170.36ª	149.39 ^a	118.45 ^a			
Size (B)														
100-150 g	42.32 ^d	602.52 ^c	128.41 ^c	250.27 ^c	239.98 ^c	149.37 ^c	11.82 ^d	13.80 ^d	33.62 ^d	55.36 ^d	53.40°			
151-250 g	66.43 ^c	684.11°	170.15 ^c	290.39 ^c	342.60 ^b	198.63 ^{bc}	21.48 ^c	32.38 ^c	93.51 ^c	89.23 ^c	72.33 ^c			
251-500 g	95.18 ^b	1059.68 ^b	396.14 ^b	431.02 ^b	388.37 ^b	224.29 ^b	31.90 ^b	50.82 ^b	129.59 ^b	151.29 ^b	155.12 ^b			
> 500 g	188.63ª	1789.91ª	695.50 ^a	842.41 ^a	707.77 ^a	426.90 ^a	50.83 ^a	93.68ª	204.88 ^a	247.53 ^a	196.18ª			
P-value														
A	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	0.001	0.813			
В	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001			
AxB	0.095	< 0.001	< 0.001	0.046	< 0.001	< 0.001	0.072	< 0.001	< 0.001	< 0.001	< 0.001			
SEM (±)	127.895	11519.650	1859.947	3583.213	2605.150	1345.800	6.833	91.455	178.914	140.097	156.706			
CV (%)	15.168	13.660	16.331	17.371	16.006	19.328	11.858	26.401	15.254	11.466	13.814			

The mineral composition of *M. cephalus* sourced from both culture and wild with different size groups is presented in Table 4. Regardless of resource and size, the highest values were found in potassium followed by phosphorus, while calcium, sodium and magnesium were in the range of 10–40 mg/100 g. The level of calcium, phosphorus, sodium and potassium in cultured fish was 18.66, 185.49, 35.76 and 231.78 mg/100 g, respectively, and was significantly (P < 0.05) increased by 10.68, 5.82, 9.31 and 6.93% in wild caught fish. The increasing trend with increasing body weight was noticed for all the four macroelements as mentioned above. However, the amount of magnesium did not vary between the source as well as size. The level of all the micro-elements analysed was < 0.5 mg/100 g and was not influenced by both resources and size.

Table 4

Mineral composition (mg/100 g wet weight basis) of different size group of *M. cephalus* collected from culture and wild resources.

Particulars	Mineral o	composition	Q I	id wild resou	1000.				
	Ca	Р	Na	K	Mg	Cu	Fe	Mn	Zn
Resource (A)	18.66 ^b	185.49 ^b	35.76 ^b	231.78 ^b	22.95 ^a	0.22 ^a	0.57 ^b	0.03 ^a	0.37 ^a
Culture	20.21 ^a	196.29ª	39.09 ^a	247.84ª	24.33 ^a	0.23 ^a	0.84 ^a	0.03 ^a	0.38 ^a
Wild									
Size (B)									
100-150 g	17.09 ^d	180.67 ^c	31.84 ^b	225.96 ^c	23.10 ^a	0.21 ^a	0.71 ^a	0.03ª	0.40 ^{ab}
151-250 g	18.40 ^c	183.49 ^{bc}	37.65 ^a	236.53 ^{bc}	23.54 ^a	0.24 ^a	0.71 ^a	0.03 ^a	0.35 ^b
251-500 g	20.60 ^b	195.80 ^{ab}	40.06 ^a	241.45 ^b	24.63 ^a	0.21 ^a	0.73 ^a	0.03 ^a	0.34 ^b
> 500 g	21.65 ^a	203.61 ^a	40.15 ^a	255.29 ^a	23.31 ^a	0.24 ^a	0.68 ^a	0.03ª	0.43 ^a
P-value									
А	< 0.001	0.030	0.009	0.001	0.066	0.312	< 0.001	0.596	0.671
В	< 0.001	0.009	< 0.001	0.001	0.436	0.217	0.908	0.541	0.031
AxB	0.746	0.508	< 0.001	0.053	0.409	0.135	0.263	0.864	0.145
		Mine							
SEM (±)	0.378	68.856	4.124	53.510	1.666	0.001	0.006	0.001	0.002
CV (%)	4.162	5.721	7.140	4.014	7.183	15.280	14.473	13.911	13.766

The daily value (DV) of lipid, cysteine, n-6 + n-3 fatty acids and calcium was < 25% and protein, valine, phosphorus and selenium were between 25–50%. Isoleucine, leucine, lysine, phenylalanine and threonine showed a range of 50–100%, whereas it crossed > 100% for methionine and EPA + DHA (Fig. 3). The DV was increased by 3.71, 3.03, 1.04, 0.78, 5.63 and 2.78% for lipid, phenylalanine, threonine, tryptophan, valine and n-3 + n-6 fatty acids, respectively, and was decreased by 0.57, 1.01, 0.45, 2.97, 7.12, 10.18, 0.26, 1.8 and 3.21% for protein, cystine, leucine, lysine, methionine, EPA + DHA, calcium, phosphorus and selenium in wild caught fish compared to the cultured species, whereas no difference was noticed for isoleucine between the resources (Fig. 4). The result of DV of fish with the varied size has shown that the fish with a body weight of > 500 g had a higher DV for lipid, lysine, methionine, EPA + DHA, n-3 + n-6 fatty acids, calcium and phosphorus, while cysteine and selenium were high in 251–500 g group. The DV of protein, isoleucine, leucine, phenylalanine, threonine and tryptophan found to be high in fish with 151–250 g weight and the group of 100–150 g showed a higher value for valine.

Discussion

Nutrient composition of fish, collected from wild and culture, is greatly varied in general. Because in intensive culture, fish are provided with nutrient dense compounded feeds that enables them to deposit large reserves of nutrients, in particular lipids. In contrast, considerable changes occurred in environmental condition fluctuates the availability and composition food that would affect the nutrient composition of wild fish²⁰. In addition to food/feed, Piggott and Tucker²¹ listed some other factors influencing the nutrient composition of fish such as species, genetics, size, reproductive status and environmental characteristics. In our study, the moisture content of wild fish was observed to be the highest compared to the cultured ones and was drastically reduced with increasing body size. This result is in agreement with the findings of Alasalvar et al², who found higher moisture content in the wild caught sea bass than the cultured fish. In the present study, muscle protein content of *M. cephalus* did not vary between the resources. Similarly, size variation had no effect on the protein level. This result is corroborated with the finding of Nettlon and Exler²² in channel catfish, coho salmon and rainbow trout. In contrast, a slight variation in protein content was observed between wild and cultured yellow perch¹⁷. The difference in protein content might be due to the variation in environmental conditions, species, size, sex of the individual animals and their reproductive status¹⁴.

In the present study, the muscle tissue of the cultured M. cephalus contained higher lipid content than the wild caught fish. This is in agreement with earlier reports that have shown that the level of lipid tend to be lower in fish collected from wild compared to cultured individuals of the same species, such as yellow perch¹⁷, turbot²³, sea bream²⁴, and silver pomfret²⁵. The difference in muscle lipid content is most likely due to the result of dietary differences¹⁷. Grigorakis et al²⁶ stated that the higher energy consumption would also be a reason for the storage of high lipid in cultured fish than the wild ones. The reduced activity of cultured fish than the wild fish would also influence the body lipid2. In addition, possible periods of starvation that encountered by wild fish might lead to reduce the lipid content of their body, which is not much occurred with cultured fish²⁷. Morishita et al¹⁵ observed a higher muscle lipid content in cultured sea bream with increasing body weight and the same is corroborated in the present investigation, indicating that this as an intrinsic physiological trait. Rheman et al²⁸ suggested that this would also be related to reproductive physiology of the fish. A similar observation has been made in sea bass²⁹ (Poli et al., 2001). But in contrast, Giogios et al³⁰ observed a lower lipid with higher body size and a higher lipid with a lower body size of cultured meagre fish. This difference would be attributed to a variation in lipid metabolism and feed offered during the culture. In most of the earlier studies 2,17,23,24, an inverse relationship was observed between the moisture and lipid content, while Zhao et al²⁵ did not find the same relationship in silver pomfret and stated that this might be because of silver pomfret is a non-fatty fish and the sample tested by the authors was originated by pooling five fishes together. Though, M. cephalus, used in the present study, is also considered as a non-fatty fish and though the analysis was done by pooling six fishes together in the present investigation, still observed an inverse relationship. Total ash content was significantly higher in wild fish compared to the cultured fish and is corroborated with our earlier findings^{9,14}. In contrast, Alasalvar et al² and Sharma et al³¹ reported no significant difference in ash content of sea bass and rohu, respectively sourced from culture and wild. The gross energy content was greatly varied between the sources (110.12-133.03 Kcal/100 g) and size groups (107.46-145.27 Kcal/100 g), which mainly related to the lipid content of fish as evidenced in the present study by a direct correlation between lipid and energy (Table 1).

It is essential to have adequate knowledge on amino acid content of fish protein to establish its nutritional value, as the nutritional quality of protein is mainly depending on EAA. Fish protein has a greater satiety effect than other animal proteins like chicken, beef, etc., with cheaper price⁷. The depicted values of amino acids in the present study was almost resembled to those presented by Mai et al³², who compared the amino acid composition of different fin-fishes, like white sucker, burbot, black crappie, rainbow trout, walleye pike and yellow perch. Of all the EAA, leucine found to be high and was in the range of 1.57-1.64% (wet weight basis). Etzel³³ documented that leucine is mainly responsible for muscle protein synthesis and later, its therapeutic effect in various stress conditions of burn, trauma and sepsis is reported by De Bandt and Cynober³⁴. Followed by leucine, M. cephalus had higher content of lysine (1.50–1.55% wet weight basis) and is agreed by Zhao et al²⁵ in Pomfret, Igtidar et al³⁵ reported that lysine is one of the most limiting amino acids in cereal-based diets given to children and is extensively required for optimal growth, hence it is suggested fish as an optimal supplement for cereal-based diets. Methionine plays a vital role in treating Parkinson's disease, liver disorder and schizophrenia, which would also help in the treatment of alcoholism, allergies, asthma, drug withdrawal, poisoning, particularly eliminating copper, radiation side effects, etc., 14 and was ranged from 0.74–0.81% in M. cephalus. Arginine was the second highest EAA in the muscle portion of M. cephalus, which is mainly considered as a precursor for the biological synthesis of nitric acid and play an important role in neurotransmission, blood clotting and blood pressure maintenance. The disease of sepsis, preeclampsia, erectile dysfunction and anxiety was much recovered with the supplementation of arginine. Isoleucine, phenylalanine, threonine and valine had a variety of roles in human nutrition mainly in chronic renal failure and nervous system disorders²⁶ and were found to be around 1% in both wild and cultured M. cephalus. Liao et al³⁷ documented the necessity of histidine in the growth and repair of tissues, myelin sheaths maintenance and to remove heavy metals from the body. Similarly, tryptophan acts as a precursor for different neurotransmitters like serotonin, dopamine and nor-dopamine and was in the range of 0.28-0.29% in our study.

Though NAA are synthesized *de nova*, they would also play a crucial role metabolically as in EAAs, especially in the regulatory process of gene expression, micro-RNA levels, metabolism, innate and cell mediated response³⁸. Deutz et al³⁹ reported that during critical illness, the effluxed glutamic acid from muscle serves as an important carrier of nitrogen as ammonia in the splanchnic area and immune system, which alone contributed more than 30% in total of NAA in our study. However, no significant difference was observed in muscle composition of both EAA and NAA, indicating that the protein content in *M. cephalus* was well balanced in amino acid composition, in particular EAA and is of high quality, irrespective of the resources and size. This result is corroborated with the findings of Gonzalez et al¹⁷ for the EAA content of wild and cultured yellow perch, but not for NAA and who reported significantly higher values for alanine, serine and tyrosine in wild caught yellow perch. Contrary, Zhao et al²⁵ reported significant differences for all the EAA except arginine between wild and cultured silver pomfret and who found lower values in cultured fish than the wild ones. In relation to identifying the quality of protein, most of the studies are restricted in analysing only EAA. But in the present study, IP level was computed based on the FAO/WHO/UNU¹⁹ recommended values to elucidate that *M. cephalus* is how far better in providing protein/amino acid content in the human diet. It was found that IP level for all the calculated EAA was higher in *M. cephalus* compared to the recommended values except valine, indicating that *M. cephalus* is of good source in providing protein/amino acids irrespective of the resources even at varying size. Sarma et al⁴⁰ found a marginal decrease in the IP value for histidine, leucine and threonine in golden mahseer, common snow trout and common carp, respectively as compared with the recommended level. Though the IP level of valine reduced than the recommended level in o

As a source of membrane constituents, energy, metabolic and signaling mediators, fatty acids, particularly n-3 PUFAs are recognized as essential nutrients for life. Aquatic species, primarily fish are generally characterized by high level of n-3 PUFAs, however, its composition influenced by many intrinsic and extrinsic factors. In our study, the level of saturated fatty acid (SFA) and monounsaturated fatty acid (MUFA) was significantly high in cultured M. cephalus than in wild caught fish, while the reverse was true for PUFA except C18:2c and yC18:3 and as reflected in n-6/n-3 ratio, which shown that significantly lower n-6/n-3 ratio in wild compared to cultured fish, indicating that marine environment would have an excellent source for n-3 rich foods and which might be lower in intensive culture system. This result is greatly in agreement with those previously reported by Zhao et al4. In contrast, Alasalvar et al2 found the reverse trend in sea bass, who reported about 29.2 and 33.4% of SFA in cultured and wild collected sea bass and the remaining quantity was shared by MUFA and PUFA. But in the present study, SFA was about 50.7% in cultured fish and almost similar level was observed for wild fish (49.6%). C16:0 was about > 70% of the total SFA content of both cultured and wild M. cephalus and was significantly higher in cultured than in wild fish, which might be attributed to the usage of supplementary feed containing higher C16:0. Similar results have also been reported in sea bass², crappie¹⁶, rohu³¹ and sturgeon⁴¹. In MUFA, C18:1 was found as a major one in the both the fish and was significantly higher in cultured fish than that of wild ones. Similarly, C18:2c found to be ten-times higher in cultured M. cephalus compared to its wild counterpart, which is due to its dominance in compounded feed used for intensive culture²⁶. Among n-3 series, both EPA and DHA had no much variation like other fatty acids between the wild caught and cultured fish, indicating that both the fish are of good sources for these fatty acids. However, a marginal increase was noticed in wild fish than the cultured ones, but the difference was insignificant for DHA. A great variation was noticed in C20:4 and was much higher in wild fish (170.36 mg/100 g) than in cultured ones (60.44 mg/100 g). A similar result was found by Gonzalez et al¹⁷ in yellow perch and who suggested that this might be due to the dietary effect and saturation and/or elongation mechanisms. Similarly, the highest concentration of aC18:3 was observed in wild fish than in culture, which could be attributed to the type of food, in which M. cephalus is exposed in the wild such as insect larvae, algae, crustacean that are rich in αC18:3⁴².

Simopoulos⁸ documented that the ratio of 1–2:1 for n-6/n-3 fatty acids is being considered as an ideal level for beneficial health, while the Department of Health of United Kingdom recommends that this level might be up to 4:1. Whereas the Western diets provide n-6/n-3 ratio of around 15–25:1, which would be a reason for occurring various common health disorders like coronary heart disease and cancer. McDanniel et al⁴³ suggested an approach, whereby consuming a higher dietary quantity of n-3 PUFAs, in particular C20:5 and C22:6 would be helpful in normalizing n-6/n-3 ratio. In our study, this ratio was 2.21:1 in cultured fish and was significantly low in wild caught fish (0.76:1), while it was in the range of 1.35–1.81:1 among the fish containing different body weight. All the values in our study were within the recommended values, indicating that *M. cephalus* could be considered as an optimal food source. Similarly, the DHA/EPA ratio seems to be lower in wild caught fish compared to cultured fish and the same was found in halibut fish⁴⁴, sea bream⁴⁵ and cat fish and tilapia⁴⁶. In our study, all the fatty acids were gradually increased with increasing body size. However, the rate of increasing was not same among all the size groups. The proportion of the increase was almost similar between 100–150 g and 151–250 g groups as well as between 151–250 g and 250–500 g groups, while the increase was almost doubled in the fish categorized under > 500 g. The similar variations in fatty acids in relation to fish size have been reported in gold-spot mullet¹⁴, rainbow trout⁴⁷ and gilthead sea bream⁴⁸. Kiessling and Kiessling⁴⁹ suggested that the relative variations on fatty acid composition with varied body weights could be attributed to the selective aerobic phosphorylation of fatty acids into the mitochondria of muscle tissue. The selective mobilisation of fatty acids to the reproductive organs in larger fish would also be a reason for the same⁵⁰. In contrast, Ghomi and Nikoo⁵¹ observed higher leve

Mineral content of fish is influenced by both diet and environment, especially in intensive culture and would also have an influence on flavor. In addition, deficiency of essential minerals in the diet might lead to improper enzyme-mediated metabolic functions, organ malfunctions and chronic disease. In our study, all the macro elements differed among the treatments except magnesium. Wild caught *M. cephalus* contained higher level of calcium, phosphorus, sodium and potassium than the cultured fish, while no significant difference was noticed for magnesium. Contrary, Gonzalez et al¹⁷ reported a higher concentration of magnesium, phosphorus, calcium and potassium in cultured yellow perch than that of its wild counterpart and the reverse was true for sodium. Whereas, micro elements were not affected except for iron. This result is agreed by Alasalvar et al² in sea bass, however, a higher level of zinc was reported in cultured eel⁵². A linear relationship was observed in macro elements against fish body weight. Environmental conditions like water chemistry, salinity, temperature and contaminants would also responsible for the change in mineral composition². Indian Council of Medical Research farms a simple recommended daily allowance quideline based on the FAO/WHO/UNU¹⁹ quideline to calculate the daily value (DV%) of food. Dayal et al⁵³ reported that food

with > 70% DV rated as outstanding and those had 50–70, 25–50, 10–25% are categorized as excellent, very good and good, respectively, while if it falls < 10% is considered as poor. According to this, lysine, methionine, threonine, tryptophan and EPA+DHA were considered as outstanding nutrients in both wild and cultured *M. cephalus*. Similarly, isoleucine, leucine, phenylalanine and valine were grouped under the excellent category. Protein, phosphorus and selenium were categorized as very good. A similar trend was noticed among the size groups. Cultured *M. cephalus* contained a higher DV for lipid, phenylalanine, threonine, tryptophan, valine and n-3 + n-6 fatty acids, whereas protein, cysteine, leucine, lysine, methionine, EPA + DHA, phosphorus and selenium were high in wild caught *M. cephalus*.

In conclusion, muscle of both cultured and wild *M. cephalus* is of good sources of protein with balanced amino acids, in particular EAA irrespective of the size groups. However, an important difference is found in some of the quality of lipid, fatty acid and macro mineral composition and they are influenced by both resources and size. In addition, the results of the present study indicate that *M. cephalus* contains higher ideal protein compared to FAO/WHO/UNU recommended level with a good daily value for most of the nutrients. Our results suggest that despite of the changes in nutrient content, both wild and cultured *M. cephalus* fall under the category of nutrient rich fish and would be an effective and healthy food material.

Methods

Different size group (100-150 g, 151-250 g, 251-500 g and > 500 g) of M. cephalus specimens of cultured and wild were sourced from brackishwater fish farm at Nagavalanka (15.9450 N 80.9180 E; Diviseema region) and Krishna River, respectively of Krishna district, Andhra Pradesh, India, Handling of animals in the present study complied with the current animal welfare laws in India and was in accordance with the relevant guidelines and regulations of the CPCSEA (Committee for the Purpose of Control and Supervision of Experiments on Animals), Ministry of Fisheries, Animal Husbandry and Dairying, Govt. of India, on care and use of animals in scientific research. This study was undertaken with approval of the Institute Animal Ethics Committee (IAEC), ICAR-Central Institute of Brackishwater Aquaculture, Chennai, India (F.No. SPA/Dir/5-90. Dated: 07.04.2014). Each group has been designed to contain six fishes with three replications. After dressed, skin and bone were removed from all the fishes. Boneless muscle tissues were collected from both dorsal and ventral areas and stored at -20° C until analysis. Proximate composition of was determined by AOAC⁵⁴ methods. The gross energy of samples was tested using a semi bomb calorimeter (Parr-1425). Amino acid profiles were analysed using pre-column HPLC gradient system (Shimadzu Corp, LC-30AD) after hydrolysing the samples with 6 N hydrochloric acid in a sealed tube filled with nitrogen for 22 h at 110° C in a vacuum oven⁵⁵. Individual amino acids were separated by YMC-Triart C18, RRH (1.8 µm, 2.1 × 100 mm) column after derivatization with mercaptopropionic acid, O-phthalaldehyde and fluorenylmethoxycarbonyl chloride. A gradient elution using phosphate buffer (20 mmol as mobile phase A) and combination of acetonitrile:methanol:water (45:40:15 as mobile phase B) at the flow rate of 0.3 ml/min was used. The gradient was changed by increasing mobile phase B concentration at the rate of 11-13% at 3 min, 31% at 5 min, 37% at 15 min, 70% at 20 min and 100% at 25 min. Amino acids were qualified and quantified by a fluorescent detector (RF-20AXS) using the amino acid mixer as an external standard (Sigma Aldrich, Cat. No: AAS18) and nor-leucine as an internal standard. Tryptophan, being labile to acid hydrolysis, was measured after alkali hydrolysis using a spectrophotometric method at 500 nm⁵⁶. Essential amino acid index (EAAI) was calculated based on the ideal protein/amino acid level recommended by FAO/WHO/UNU¹⁹.

Lipid was extracted using a combination of organic solvent (chloroform and methanol at 2:1 ratio) and the process of saponification followed by esterification was done according to Metcalfe et al 57 . FAMEs were subsequently analysed using a gas chromatograph (GC-2014 Shimadzu) on a RTX-Wax Capillary Column (100 m length × 0.25 mm l.D × 0.2 μ m film thickness). Nitrogen was used as the carrier gas at a linear velocity of 20.9 cm/s with 3 ml/min of purge flow. The oven temperature was held at 100° C for 4 min and increased to 225° C at the rate of 3° C/min and held for 5 min and further increased to 240° C at the rate of 1° C/min. The operating temperature for injection ports and flame ionization detector was 225 and 250°C respectively. Individual fatty acids were identified by comparing with the retention times of 37 Component FAME Mix (Supelco-Sigma). Tridecanoic acid methyl ester (C13:0, Supelco-Sigma, USA) was used as an internal standard to calculate fatty acid content in the sample (mg/100 g). Mineral composition was estimated according to the method of Jannathulla et al 58 . Briefly, a gram of sample was digested with 6 ml of HNo $_3$ and 2 ml of H $_2$ O $_2$ in microwave digestion system (Anton Paar). All digested samples were analysed in triplicates using inductively coupled plasma-optical emission spectrometry (ICP-OES) in an instrument of Agilent-5100, using the 5.2 software. The analytical measurements were made with an autosampler equipped with a peristaltic pump, across-flow nebulizer (coupled to a double-pass spray chamber) and Quartz central torch tube injector with an internal diameter of 2 mm. Certified reference material, ICP multi-element standard solution (10 mg/l Merck), was used for calibration.

The statistical package of SPSS ver.16.0 for Windows was used to assess the significant differences between the means of experimental data according to a two-way analysis of variance (ANOVA) using a 2×4 factorial design with two different factors such as resources (culture and wild) and size (100-150 g, 151-250 g, 251-500 g and 500 g). The descriptive statistical measures were calculated for the two main factors and their interactions. For this, post-hoc was done with Tukey's test and differences were considered significant when P < 0.05. Regression analysis was performed to find a correlation on EAAI between the resources irrespective of the size groups. Prior to statistical evaluation, data were checked for homogeneity of variances after ascertaining normal distribution.

Declarations

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Authors contributions

J.S.D. was responsible for the study design and written the manuscript, R.J. analyzed all the parameters, K.A. did statistical analysis, K.P.K.V. helped for analysis and to write this manuscript, K.K.V. reviewed the data and the manuscript.

Competing interest

The authors declare no competing interest.

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Figures

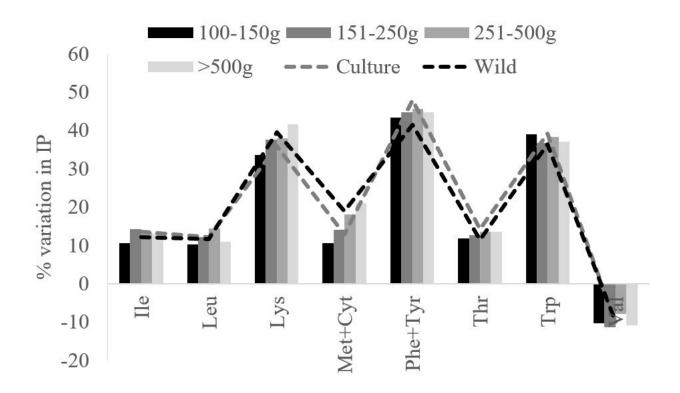


Figure 1

Percent variation of ideal protein (IP) level of M. cephalus collected from different resources with varied size groups.

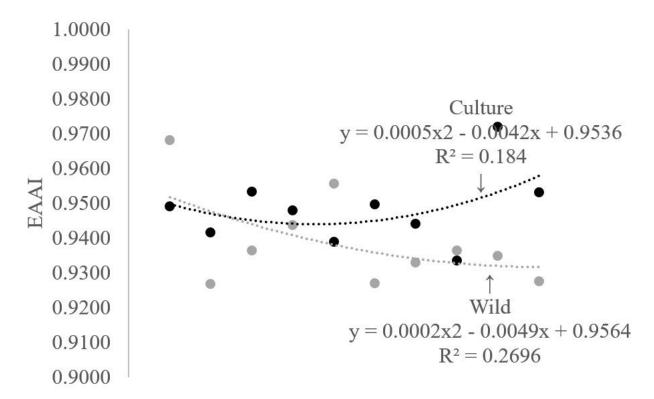


Figure 2

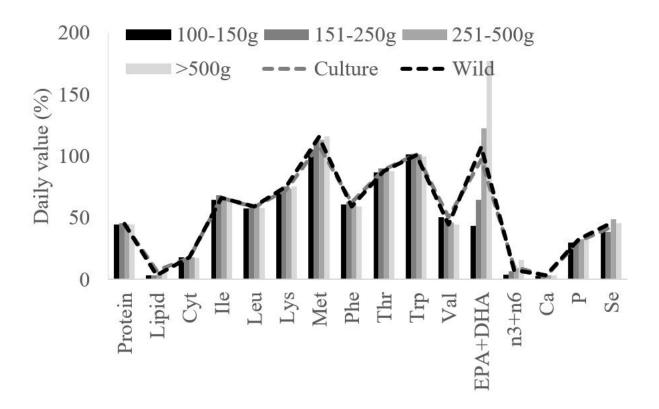
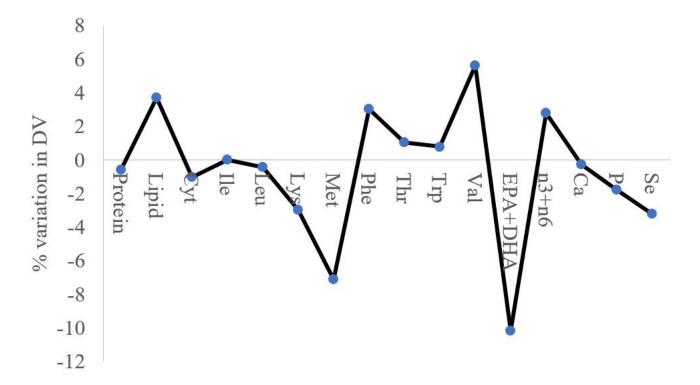


Figure 3

Daily value (%) of one serving of 100g of M. cephalus collected from different resources with varied size groups.



Percent variation of daily value (DV) between culture and wild collected M. cephalus irrespective of the size groups.

Figure 4