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6. Nutritional Status of Small Indigenous Food Fish in Terms of Their Proximate Composition, Amino Acid, Fatty Acid, Vitamin and Mineral Contents

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Abstract:

The nutrient qualities of small fishes are a global point of research for the sustainable development of the nation. Indian spiny eel (Macrognathus pancalus) and Bothia (Noemacheilus corica) are two commonly available small fish widely consumed by the different communities of Kokrajhar, BTAD, Assam. These fish species are found in weedy bodies of waterlogged areas. The present study was undertaken to estimate the proximate composition, amino acid profile, fatty acids composition and selected minerals and vitamins of the small fish species. It is reported that the studied fish have significant moisture, ash, protein carbohydrate and crude lipid contents. The amino acid profile of the fish protein showed the higher content of the few essential amino acids. The fish lipid was analyzed to sort out the fatty acids and the result highlighted the presence of all the poly unsaturated fatty acids in small amounts. Analysis of selected vitamins and minerals concluded the higher content of calcium and phosphorous. Both the fish species may be included regularly in the human diet for nutritional benefit.

Keywords:

Amino acid profile, fatty acid composition, proximate composition, minerals and vitamins.

6.1 Introduction:

In recent years due to the toxic effects ‘formalin’, a chemical that is used to preserve fishes, people are raising their concern to consume the imported fishes and in this regard the demand of small and indigenous fishes (SIFs) are growing higher. Due to their ease in availability and lower market price, these fishes are regularly included in their diet. The local people cannot afford the costly big imported fishes as well. Therefore, the SIFs can become one of major replacements of the imported fishes in human diet. A common and easily available SIF is the spiny eel or Indian spiny eel (*Macrognathus pancalus*) that is commonly known as Turi in India. The fish species are inhabited in slow and shallow water of rivers of plains and estuaries, they are never available above and altitude of 366 mm. These are also found in canals, streams, beels, ponds and inundated field (Rahman, A. K., 1989). These species are placed into aquarium by hobbists. It stays on the bottom and spauns in the upper water level (Talwar and Jhingran, 1991). Bothia (*Noemacheilus corica*) is another SIF which are commonly known as loasch fishes. These have been recorded as the member of the genus Noemacheilus. They have extended pectoral fins, a complete lateral line and colour pattern consisted of several dark spots arranged through the dorsal surface from head to caudal peduncle. Currently the genus name of the species has been replaced to Nemacheilus (Kottelate, 2012)**.** The genus Nemacheilus is having some prominent characteristics such as elongated body, complete lateral line, presence of enlarge scales in some species, large eyes, small mouth, thin lips etc. The family Nemacheilidae is largely distributed across most of Euro-Asia with the Indian subcontinent, South East Asia and China representing particular centres of species diversity.

Kingdom**:** Animalia
Phylum: Chordata
Class: Actinopterygii
Order: Synbranchiformes
Family: Mastacembelidae
Genus: Macrognathus
Scientific Name:
Macrognathus pancalus
(Hamilton, 1822)
Local Name:Turi

Figure 6.1 ***Macrognathus pancalus***

Kingdom**:** Animalia
Phylum: Chordata
Class: Actinopterygii
Order: Cypriniformes
Family: Bollidae
Genus: Botia
Scientific Name:
Noemacheilus corica
(Hamilton, 1822)
Local Name: Bothia

Figure 6.2 ***Noemacheilus corica***

The proximate analysis basically emphases on the quantification of many nutritional contents of fish. The nature and quantity of nutrients in most food fishes depends on number of factors based on their feeding habits. The flesh of fish in good condition is consisted of five main chemical components as protein, lipid, water, mineral and vitamins. All of them contribute towards the total meal composition. The proximate analysis is investigated implementing several standard methods to estimate the vital components present in fish which become a deciding factor for the nutritional status of fish. The composition of fish body is affected by both exogenous and endogenous factors (Huss 1995). The exogenous factors affect the body composition as well as the diet of the fish (composition, frequency) and also their existing environment (salinity, temperature). A number of researchers examined the influence of temperature, light, pH and the oxygen concentration on the proximate composition of fish. The endogenous factors include some genetic factors linked to the life stage, size, age, sex and anatomical position in the fish (Huss 1995). Dietery protein provide substrates needed for synthesizing body proteins and also other nitrogen-containing compounds. The amount of amino acid contents in food proteins determine the nutritional quality of protein (Young et al., 1994). Fish is an important source of mineral like Potassium, phosphorous, iron, sodium, magnesium, iodine, zinc, calcium etc. Mineral components find their vital importance for human nutrition (Love. R.M. 1957). Some of the trace elements such as iron, manganese and iodine are adequately present in fish species (Borgstrom, G. 1962). Several literatures justified that fish is a good source of vitamins like A, D, E and K. The contents of Vitamin A originated from fish food is easily demonstrated to the human body than the same from plant source (Liu, 2003). As far as the human health is concerned, Vitamin A is mainly required for normal vision and growth of the bones.

6.2 Methodology:

Total of 500 gm fish species bought from the local markets. They were immediately transported to the laboratory in ice box. In the laboratory they were thoroughly washed under tap water. The average length and breadth were measured to the nearest centimetre scale and the body weight were measured to the nearest gram. The fish samples were tightly packed in an airtight container and stored frozen until biochemical analysis.

6.2.1 Biochemical Analysis:

The moisture and ash contents of the fish species were estimated following DGHS LAB MANUAL (6.0) and the protein and carbohydrate content were estimated using the method IS: 7219:1973 (RA 2005) and IS: 1656-2007) respectively. The percentage of total lipid were estimated following DGHS Lab manual.

Amino acid composition was determined by following the method of QA.16.5.10. Analysis of Vitamin A and Vitamin D was performed by using HPLC according to the method of (QA.16.5.3). The Fatty acid profile was done using GC (FID) according to the method described by (AOAC 19 th edition 996.06). The mineral such as Iron, Zinc, Calcium were estimated by using UV- Visible Spectrophotometer, AAS, according to the method of (QA.16.5.2). Phosphorus was estimated by using uv-visible spectrophotometer, AAS following the method of (IS:14828:2000).

6.3 Results and Discussion:

6.3.1 Proximate Composition:

The proximate composition of all the fish species selected for the study are shown in Table:6.1 below. It is found that, the species *Noemacheilus corica* contain higher amount of protein and moisture, while the species *Macrognathus pancalus* contain higher amount of crude lipid, ash and carbohydrates per 100 g of the samples. The protein contents of both the fish species did not differ too much which may be due to their similar absorption capability and conversion potentials of essential nutrients from the local environment (Adewole et. al., 2003).

The protein content in the fishes indicated that small food fishes are beneficial for meeting our daily requirements of protein provided they are having a balanced amino acid. The adults, pregnant and lactating mothers requires 24-56 g, 13-19 g and 71 g proteins respectively per day (Anon, 2002).

Table 6.1 Proximate Composition of the selected fish species

Fish species	Protein (g/100g)	Crude Lipid(g/100g)	Moisture(g/100g)	Ash(g/100g)	Carbohydrate (g/100g)
Noemacheilus corica	16.72±0.02	3.42±0.02	76.71±0.01	2.85±0.03	0.40±0.06
Macrognathus pancalus	14.26±0.02	5.08±0.04	70.96±0.02	5.79±0.04	3.91±0.04

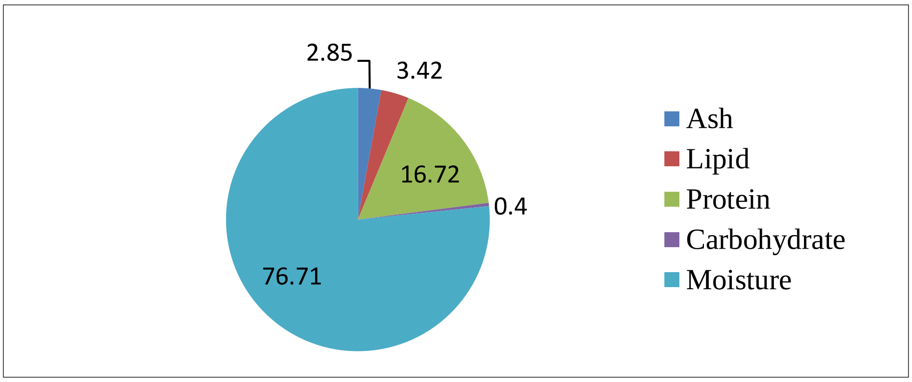

Figure 6.3: Proximate composition (%) of *Noemacheilus corica*

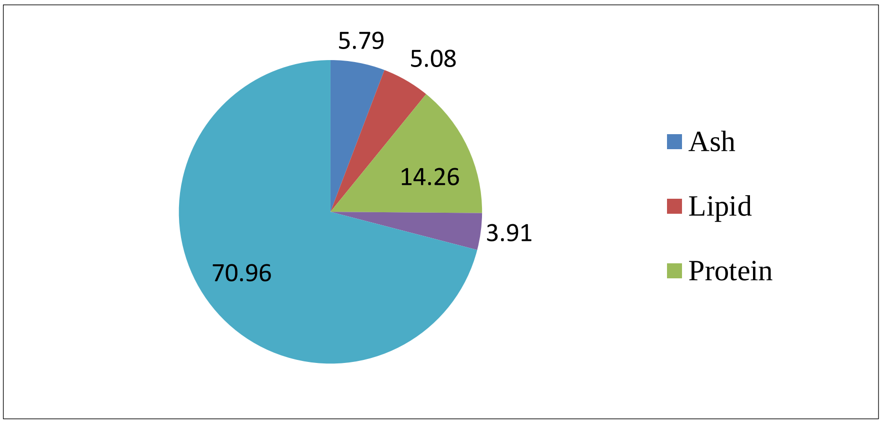

Figure 6.4: Proximate composition (%) of *Macrognathus pancalus*

According to the study of Mazumder et al. 2019 the fish moisture steadily decreases with increase in body lipid. As per the report of Marichamy et al 2012, the value of moisture content of marine fishes averaged (74%). Generally freshwater fishes were experimentally established to have low lipid content (Zhang et al. 2014) and high moisture contents. Present study revealed the finding of previous researchers to highlight good moisture contents of all the selected small fishes for study. The moisture contents of the selected fishes were negatively correlated with the lipid contents and this fact was agreed with many previous researches (Begard et al, 2015). The ash content of *Macrognathus pancalus* (Turi) fish revealed fair amount which supported FAO (2005) report. Ash contents reflect the mineral concentration present in the fish species (Ayanda et al., 2019, FAO, 2012). The poor contents of carbohydrates in the fishes may be attributed to the fact that glycogen has not much contribution to the reserves in the tissues of fish body (Das et al., 2001; Jayasre et al., 1994). There are very few fish species which are having higher values of carbohydrate contents.

6.3.2 Amino Acid Profile:

The fish species were reported to be rich in amino acid like L-Arginine (3.76 g/100g) and L-Methionine (6.80 g/100g) in *Noemacheilus corica* and L-Asparagine (1.59 g/100g) and L-Threonine (7.07 g/100g) in *Macrognathus pancalus,* as shown in Table.4(a,b,c) . L-Glutamic acid, L-Isoleucine, L-Leucine and L-Valine are present in both species to some extent. L-Histidine (0.23 g/100g) is an important amino acid which is present in *Noemacheilus corica* only. Histidine plays multiple roles in protein interactions (Liao et al.,2013) and good precursor of histamine. It is required for the growth and repair of tissue, maintenance of myelin sheath, removing heavy metals from the body (Heimann, 1982). The fish species (both) may be recommended for the daily consumption to enrich the human nutrition. Previous researchers documented the high contents of Histidine in another small indigenous fishes like *A. testutidineous*, *A. mola* and *P. sophore* (A. Mahanty et al.,2014). In the present study the some of the amino acids like L-Alanine, L-Aspartic acid, L-Cystine, L-Glutamine, L-Glycine, L-Proline, L-Serine, L-Tyrosine, L-Isoleucine, L-Leucine, L-Phenylalanine, L-Threonine, L-Tryptophan were detected in smaller extent. All of them are daily required by human for healthy living (Joint FAO/WHO/UNO 2007). An essential amino acid Methionine was highly quantified in *Noemacheilus corica*.

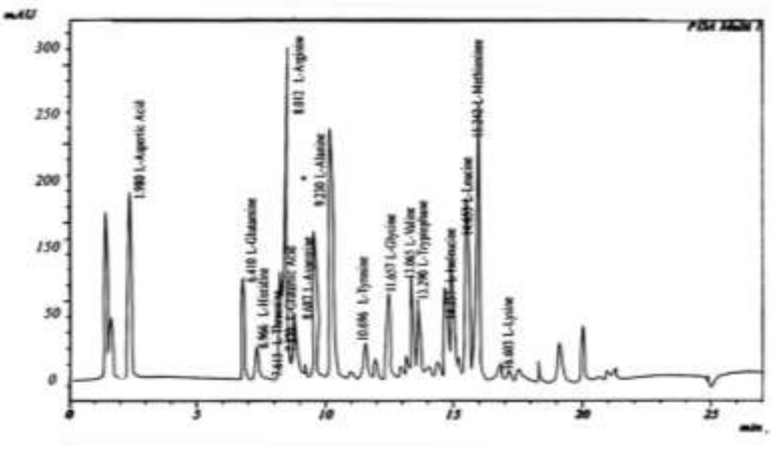

Figure 6.5: HPLC Chromatogram of Amino acids of *Noemacheilus corica*

Table: 6.2 Peak Table for HPLC Data of amino acid of *Noemacheilus corica*

Peak #	Name	Area	Area %
1	L-Alanine	880423.8	1.999
2	L-Arginine	5610837	12.741
3	L-Asperginine	149224.4	0.339
4	L-Aspartic acid	3148634	7.150
5	L-Glutamic acid	268603.9	0.610
6	L-Glutamine	1149028	2.609
7	L-Glycine	104457.1	0.237
8	L-Histidine	343216.1	0.779
9	L-Isoleucene	19399169	44.053
10	L-Leucine	850579	1.932
11	L-Methionine	10147258	23.043
12	L-Tryptophan	940113.6	2.135
13	L-Valine	1044571	2.372
Total		44036114	100.000

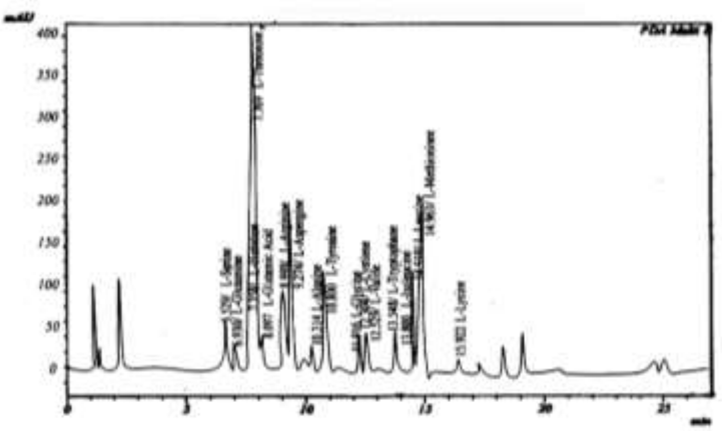

Figure 6.6: HPLC Chromatogram of Amino acids of *Macrognathus pancalus*

Table 6.3: Peak Table for HPLC Data of amino acid of *Macrognathus pancalus*

Peaks #	Name	Area	Area%
1	L-Serine	514099	4.149
2	L- Glutamine	89692	0.724
3	L- Histidine	2728	0.022
4	L- Threonine	4239364	34.210
5	L- Giutamic Acid	227660	1.837

Peaks #	Name	Area	Area%
6	L- Arginine	702832	5.672
7	L-Aspergine	1766264	14.253
8	L-Alanin	186753	1.507
9	L- Tyrosine	774386	6.249
10	L- Glycine	5530	0.045
11	L- Cystiene	401298	3.283
12	L- Valine	282856	2.283
13	L-Trytophane	58992	0.476
14	L- phenylanine	360275	2.907
15	L- Isoleucine	251355	2.028
16	L- leucine	1000536	8.074
17	L-Methionine	1514883	12.225
18	L- Lysine	12533	0.101
Total		12392037	100.000

Table 6.4a: Amino acid profile of the selected fish species

Fish species	L-Alanine g/100g	L-Arginine g/100g	L-Asparagine g/100g	L-Aspartic acid g/100g	L-Cystine g/100g	L-Glutamic acid g/100g	L-Glutamine g/100g	L-Glycine g/100g	L-Proline g/100g	L-Serine g/100g
Noemacheilus corica	0.59	3.76	0.10	2.11	BDL	0.18	0.77	0.07	BDL	BDL
Macrognathus pancalus	0.09	1.33	1.59	BDL	1.19	0.16	BDL	BDL	0.11	0.41

Table 6.4b: Amino acid profile of the selected fish species

Fish species	L-Tyrosine(g/100g)	L-Histidine(g/100g)	L-Isoleucine(g/100g)	L-Leucine(g/100g)	L-Lysine(g/100g)	L-Methionine(g/100g)	L-Phenylalanine(g/100g)	L-Threonine(g/100g)	L-Tryptophan(g/100g)	L-Valine(g/100g)
Noemacheilus corica	BDL	0.23	0.13	0.57	BDL	6.80	BDL	BDL	0.63	0.70
Macrognathus pancalus	0.75	BDL	0.19	0.73	BDL	1.03	0.46	7.07	BDL	1.03

Table 6.5a: Fatty acid profile of the selected fish species. (UOM=g/100g)

Fish species	Undecan-oic acid	Lauric acid	Tridecan-oic acid	Myristic acid	Myristoleic acid	Pentadecanoic acid	Palmitic acid	Palmitoleic acid	Heptadecan-oic acid	Heptadecen-oic acid
Noemacheilus corica	0.03	0.16	BDL	0.17	0.03	0.09	1.08	0.09	0.17	0.04
Macrognathus pancalus	BDL	0.18	0.13	0.13	0.31	0.12	1.35	0.44	0.14	BDL

Table:6.5b: Fatty acid profile of the selected fish species

Fish species	Stearic acid	Oleic acid	Elaidic acid	Linoleic acid	Linolelaidic acid	ɣ-linolenic acid	α-linolenic acid	Arachidic acid	Cis -11-eicosenoic acid	Cis-11,14-eicosadienoic acid
Noemacheilus corica	0.48	0.66	0.04	0.09	0.02	BDL	0.06	0.02	0.07	0.01
Macrognathus pancalus	0.44	1.26	BDL	0.13	BDL	BDL	BDL	BDL	BDL	BDL

Table: 6.5: c Fatty acid profile of the selected fish species.

Fish species	Cis-8,11,14-eicosatrienoic acid	Cis-11,14,17-eicosatrienoic acid	Arachid-onic acid	Cis-5,8,11,14,17-eicosapentaenoic acid	Behenic acid	Erucic acid	Cis-13,16-docosahexaenoic acid	Cis-4,7,10,13,16,19-docosahexaenoic acid	Tricosan-oic acid	Nervonic acid
Noemacheilus corica	0.01	BDL	0.04	0.02	0.02	0.007	BDL	0.005	0.007	BDL
Macrognathus pancalus	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL

BDL=Below Detectable Limit

6.3.3 Fatty Acid Profile:

The fatty acid contents of the selected fish species are shown in Table 6.5 (a,b,c). The present investigation recorded the fatty acid compositions of all the fish species. The fatty acids were not present in higher contents in the studied fishes. Although in lower amounts palmitic acid was detected in both the species *Noemacheilus corica* (1.08 g/100 g) and *Macrognathus pancalus* (1.35 g/100 g) respectively. Fair values and present in all the fish species oleic acid, stearic acid and Linoleic acid were present in both species but in lower contents. A total number of 39 individual fatty acids had been analysed for the selected small fish species. Amongest them palmitic acid was predominantly detected in all the fish species. It was followed by oleic acid, stearic acid and Linoleic acid. The present study indicated that the small fish species cannot be regarded as highly fatty fishes. The present work confirms that the selected fish species contained lower amount of Omega-6 & Omega-3 fatty acids. Harper et al., 2001 studied that a lower proportion of Omega-6 to Omega-3 fatty acids is appreciated in reducing the risk of Chronic disease in the developing countries. In this context, it is attributed that all the small fish species could be recommended safely for human consumption. Simopoulos, 2008 further studied that a balanced n-6: n-3 PUFA ratio is widely accepted in the prevention of cardiovascular disease. This fact agreed to higher risk of life alarming problems with low intake of n-3, rather that high n-6 intake (FAO, 2010).

The presence of free fatty acids above 1.5% throws an indication of unsuitability of the lipid for the sake of edible purpose (Molla et al., 2007). The record of lower percentage of free fatty acids in the lipids of studied species might be a conclusion that the fishes are suitable for edible purpose.

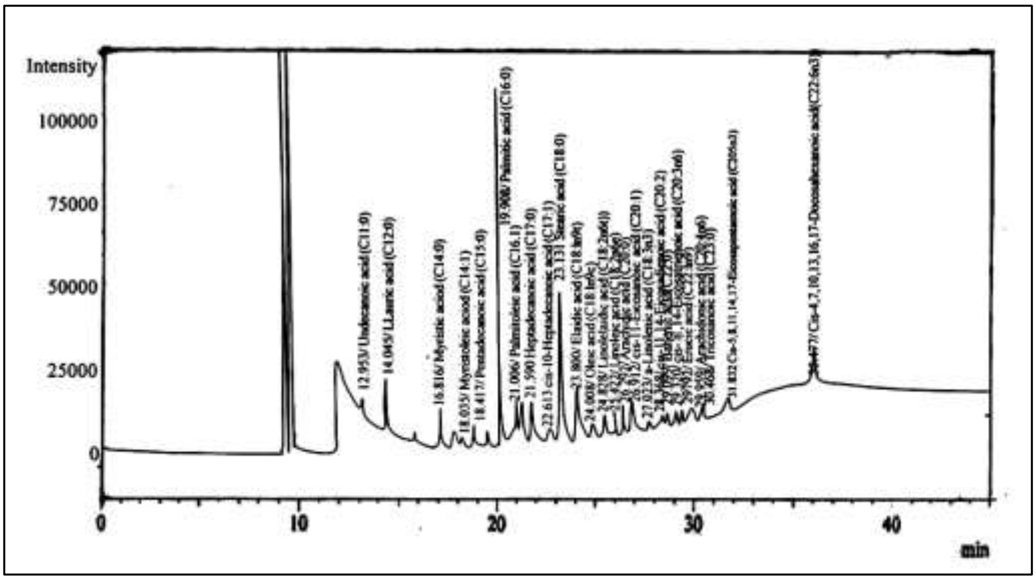

Figure 6.7: GC-MS Chromatogram of Fatty acids in *Noemacheilus corica*

Table 6.6: Peak Table Fatty acids of *Noemacheilus corica*

Peak#	Name	Ret. Time	Area	Height	Area%
1	Undecanoic acid(C11:0)	12.953	18833	2687	0.9604
2	Lauric acid(C12:0)	14.013	90372	12434	4.6086
3	Myristic acid(C14:0)	16.816	97433	30890	4.9687
4	Myristoleic acid(C14:1)	18.035	14470	1157	0.2379
5	Pentadecanoic acid(C15:0)	18.117	19192	1802	2.5239
6	Palmitic acid(C16:0)	19.908	617612	99752	31.4957
7	Palmitoleic acid(C16:1)	21.006	50963	7348	2.5989
8	Heptadecanoic acid(C17:0)	21.590	99340	10716	5.0660
9	Cis-10-Heptadecanoic acid(C17:1)	22.613	24089	2312	1.2285
10	Stearic acid(C18:0)	23.131	277120	41882	14.1320
11	Elaidic acid(C18:9nt)	23.800	24131	4826	1.2306
12	Oleic acid(C18:1n9c)	24.008	379892	40517	19.3720
13	Linolelaidic acid(C18:2n6t)	24.828	11427	1760	0.5827
14	Linoleic acid(C18:2n6c)	25.422	50091	5966	2.5544
15	Arachidic acid(C20:0)	26.292	9158	1276	0.4676
16	Cis-11,14-Eicosadienoic acid(C20:1)	26.912	37539	6963	1.9143
17	a-Linolenic acid(C18:3n3)	27.023	33815	4036	1.7244
18	Cis-11,14-Eicosadienoic acid(C20:2)	28.368	8515	1252	0.4343
19	Behenic acid(C22:0)	29.099	13268	2041	0.6766

Peak#	Name	Ret. Time	Area	Height	Area%
20	Cis-8,11,14-Eicosatrienoic acid(C20:3n6)	29.320	7793	978	0.3974
21	Erucic acid(C22:1n9)	29.792	3801	652	0.1938
22	Arachidonic acid(C20:4n6)	29.959	25259	3458	1.2881
23	Tricosanoic acid(C23:0)	30.468	4139	772	0.2111
24	Cis-5,8,13,16,17-Eicosapentaenoic acid(C20:5n3)	31.832	9485	1493	0.4837
25	Cis-4,7,10,13,16,17-Docosahexanoica acid(C22:6n3)	35.447	2921	431	0.1490
Total			1960938	270401	100.0000

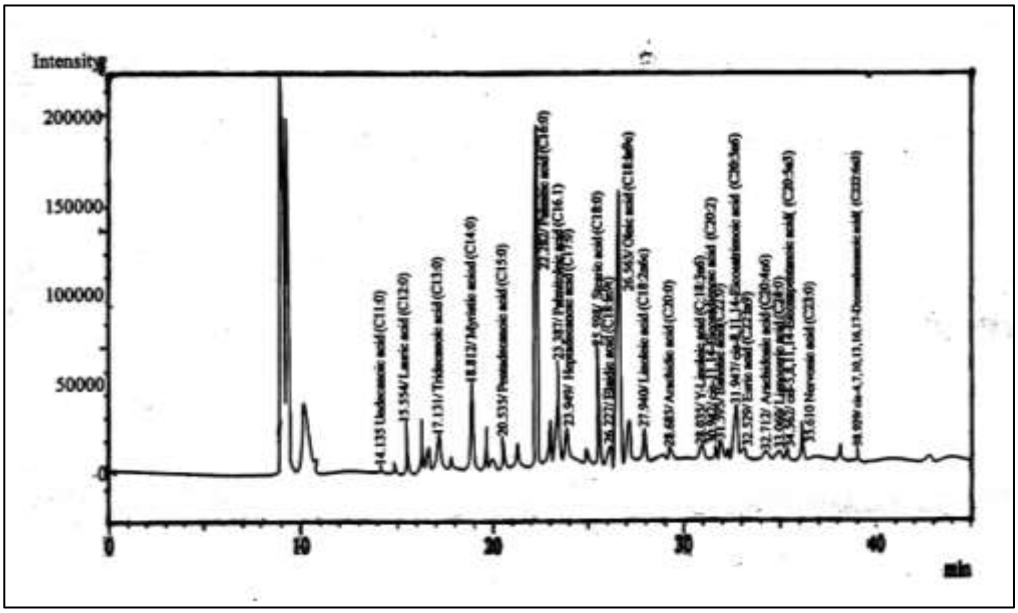

Figure 6.8: GC-MS Chromatogram of Fatty acids in *Macrognathus pancalus*

Table: 6.7 Peak Table of Fatty acids of *Macrognathus pancalus*

Peak#	Name	Ret.Time	Area	Height	Area%
1	Undecanoic acid(C11:0)	14.141	7633	2281	0.3794
2	Lauric acid(C12:0)	15.560	16225	4666	0.8064
3	Tridecanoic acid(C13:0)	17.136	22471	6521	1.1168
4	Myristic acid(C14:0)	18.815	142297	41400	7.0722
5	Pentadecanoic acid(C15:0)	20.537	94133	27371	4.6785
6	Palmitic acid(C16:0)	22.290	813944	218854	40.4534
7	Palmitoleic acid(C16:1)	23.387	121281	35525	6.0277
8	Heptadecanoic acid(C17:0)	23.950	78950	23362	3.9239

Peak#	Name	Ret.Time	Area	Height	Area%
9	Stearic acid(C18:0)	25.601	259948	76815	12.9195
10	Elaidic acid(C18:1n9t)	26.224	23917	4223	1.1887
11	Oleic acid(C18:1n9c)	26.554	351279	82297	17.4588
12	Linoleic acid(C18:2n6c)	27.940	17138	4816	0.8518
13	Arachidic acid(C20:0)	28.685	14408	4264	0.7161
14	Y-Linolenic acid(C18:3n6)	28.934	4292	1167	0.2133
15	Cis-11-Eicosenoic acid(C20:1)	29.465	5922	1284	0.2943
16	a-Linolenic acid(C18:3n3)	29.598	5558	1655	0.2762
17	Henicosanoic acid(C21:0)	30.145	1841	612	0.0915
18	Behenic acid(C22:0)	31.596	9648	2776	0.4795
19	Cis-11,14,17-Eicosatrienoic acid(C20:3n3)	32.536	2523	664	0.1254
20	Arachidonic acid(C20:4n6)	32.702	1548	395	0.0770
21	Tricosanoic acid(C23:0)	33.061	2614	773	0.1299
22	Cis-5,8,11,14,17-Eicosapentaenoic acid(C20:5n3)	34.562	9010	2062	0.4478
23	Nervonic acid(C24:1)	35.592	5472	1611	0.2719
Total			2012052	545394	100.0000

6.3.4 Mineral & Vitamin Profile:

The mineral contents of individual species are dependent on the abundance of the elements in their local enviorment, capacity of diet absorption and their accumulation as preferred (Hei and Sarojini, 2012).

The minerals paly vital roles in sketatal formarion, maintenance of colloidal systems, regulation of acid – base equilibrium. Major components of some biologically important compounds like as hormones and enzymes etc. are made of the minerals.

Different types of biochemical structural and functional pathologies are caused by mineral deficiencies (Sankar et al., 2013). Iron is having tremendous importance in the formation of hemoglobin which is essential for the formation of red blood cells (Miller, 2013). Fe is the vital element for the metabolism of almost entire living organisms.

A lot of procedures of cellular metabolism are dependent on zinc (Hei and Sarojini, 2012). The micro elements like Fe and Zn are vitally important for the formation of skeleton structure, transfer of electrons, regulation of acid-base equilibrium as well as osmoregulation. In the present investigation the iron contents were sufficiently higher in both the fish species. Higher content of iron (5.95 mg) was recorded in *Noemacheilus corica*.

The content of zinc and Calcium were also in a good amount in both the fishes. The species *Macrognathus pancalus* have higher content of phosphorous 1620 mg) was recorded. The overall mineral and vitamin content is shown in Table.6.8 and Table.6.9 respectively.

Table 6.8: Mineral (Fe, Zn, P & Ca) contents in different fish species

Scientific name of species	Fe content (mg/100g)	Zn content (mg/100g)	P content (mg/100g)	Ca content (mg/100g)
Noemacheilus corica	5.95	2.84	825.64	591.20
Macrognathus pancalus	3.75	1.44	1620.00	389.32

Table: 6.9: Vitamin A and Vitamin D content in different fish species

Scientific name of species	Vitamin A (µg/100g)	Vitamin D (µg/100g)
Noemacheilus corica	34.97	15.33
Macrognathus pancalus	111.85	BDL

Several literatures justified that fish is a good source of vitamins like A, D, E and K. The contents of Vitamin A originated from fish food is easily demonstrated to the human body than the same from plant source (Liu, 2003). As far as the human health is concerned, Vitamin A is mainly required for normal vision and growth of the bones. The specific diseases caused by the deficiency of Vitamin A include Night blindness and keratomalacia significantly in children and reproducible women (West, 2002). The present study revealed that the studied small fishes were rich source of vitamin A. The higher contents of Vitamin A (111.85 µg/100g) were found in the small fish *Macrognathus pancalus*. Vitamin D has its crucial role in regulating calcium phosphate balance in such way that stimulates calcium absorption by the small intestine and hence promoting bone metabolism. The fat-soluble vitamins are documented to be the vital nutrients which control many biologically important processes in human body. The vitamin D contents of *Noemacheilus corica* is found to be 15.33 µg/100g.

6.4 Conclusion:

The small fish species investigated in this study are experimented to be enriched in higher nutrient profile. These fishes may show excellence if commercially explored. Scientific measures should be opted for better preservation of them. They are important also from the medicinal point of views. Furthermore, studies are required on investigation of the nutritional aspects of these fishes. In our present study it may be inferred that the fish species may be a good choice for human consumption as well as research works.

Declaration: The Author declares that there is no conflict of interest.

6.5 References:

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