Journal of Food Engineering and Technology 2025;14(1):28-34
https://doi.org/10.32732/jfet.2025.14.1.28

Proximate Composition and Fatty Acid Profiling of Five Species of Fish Caught from Kerala Coast, India

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Received: 12 March 2025; Accepted: 5 June 2025; Available online: 30 June 2025

Abstract: In this comprehensive study, we investigated the proximate composition and fatty acid profiles of five distinct species of fish: *Epinephelus diacanthus*, *Johnius glaucus*, *Scatophagus argus*, *Alepes djedaba*, and *Stolephorus commersonnii*, all sourced from the vibrant waters along the Kerala coast. The proximate composition of these fish was meticulously analyzed, focusing on key nutritional components: moisture, protein, fat, and ash content. Among these species, *S. commersonnii* exhibited the highest moisture content, showcasing its fresh and succulent nature, while *A. djedaba* displayed the lowest moisture levels. In terms of protein, *S. argus* stood out with the highest concentration, indicating its potential as a rich source of this essential nutrient, whereas *E. diacanthus* came in at the lower end of the spectrum. When we examined fat content, *S. argus* again excelled with the richest levels of fat, contributing to its flavor and caloric density, while *S. commersonnii* contained the least fat, offering a leaner option for health-conscious consumers. Delving deeper into the fatty acid profiles, we classified the fatty acids into three categories: saturated fatty acids (SFA), monounsaturated fatty acids (MFA), and polyunsaturated fatty acids (PUFA). Significantly, our findings revealed that *S. commersonnii* had the highest percentage of docosahexaenoic acid (DHA), a vital n-3 fatty acid known for its numerous health benefits, while *S. argus* recorded the lowest DHA level. On the other hand, eicosapentaenoic acid (EPA) was most abundant in *Alepes djedaba* presenting an excellent option for consumers seeking the benefits of n-3s, with *Scatophagus argus* trailing behind in EPA content. Furthermore, the analysis highlighted the impressive levels of polyunsaturated fatty acids in *Stolephorus commersonnii*, making it a standout choice for those looking to enhance their diet with essential fats. Overall, this study enhances our understanding of the nutritional profiles of these fish species, providing valuable insights for both consumers and the fishing industry.
Keywords: Nutrient profiling; Eicosapentaenoic acid; Docosahexaenoic acid.

1. Introduction

Fish hold an essential position in diets globally, offering a rich array of high-quality proteins, fats, vitamins, and minerals. Remarkably, around 80% of the animal protein consumed worldwide is derived from fish and fish products [1]. The inclusion of fish in a nutritious and balanced diet is linked to a wealth of health benefits, making it a key dietary component. Packed with high-quality protein, fish is also a prime source of long-chain n-3 polyunsaturated fatty acids (n-3 PUFAs), as well as vital micronutrients such as selenium, iodine, potassium, vitamin D, and B vitamins [2] and also Fish meat contains a variety of nutrients beneficial to humans, such as carbohydrates, lipids, amino acids, trimethylamine oxide (TMAO), and more [3]. Fish proteins contain high levels of essential amino acids, especially methionine and lysine, unlike most plant-based proteins, which are often deficient in one or more essential amino acids [4]. Numerous studies have demonstrated that eating fish can reduce the risk of cardiovascular diseases, obesity, inflammation, diabetes, and cancer, due to its rich content of polyunsaturated fatty acids (PUFA), essential minerals, and high-quality protein [5]. Accordingly, the regular consumption of fish, preferably at least twice per week, is strongly recommended due to its associated health benefits [6]. To evaluate the nutritional profile of fish, researchers meticulously analyze their proximate composition, which encompasses moisture, lipids, proteins, and ash content. The composition of fish generally includes 2–25% fat, 15–30% protein, and 50–80% moisture, depending on the species and other factors [7]. Furthermore, the analysis delves into the fatty acid content to unveil the presence of n-3 fatty acids, such as DHA and EPA, which are predominantly sourced from various fish species. In this study, five remarkable fish species were investigated for their proximate composition and fatty acid profiles: *Epinephelus diacanthus* (spinycheek grouper; Fig. 1a), *Johnius glaucus* (Pale spotfin croaker; Fig. 1b), *Scatophagus argus* (spotted scat; Fig. 1c), *Alepes djedaba* (shrimp scad; Fig. 1d), and *Stolephorus commersonnii* (Commerson's anchovy; Fig. 1e). *E. diacanthus*, with its striking appearance, is a commercially significant species typically found over muddy sand or mud substrates, and is usually caught at depths ranging from 63 to 100 meters off the scenic Kerala coast. *J. glaucus*, known for its adaptability, roams the shallow, muddy coastal waters of the western Indian Ocean. *S. argus*, revered in traditional Chinese medicine, thrives in diverse environments such as brackish waters, estuaries, and marine habitats, demonstrating its versatility. *A. djedaba* is a fascinating marine, reef-associated amphidromous fish, showcasing remarkable behavioral adaptations, while *S. commersonnii* inhabits the vibrant and biodiverse tropical coastal marine and estuarine waters, contributing to the rich tapestry of aquatic life in these regions.

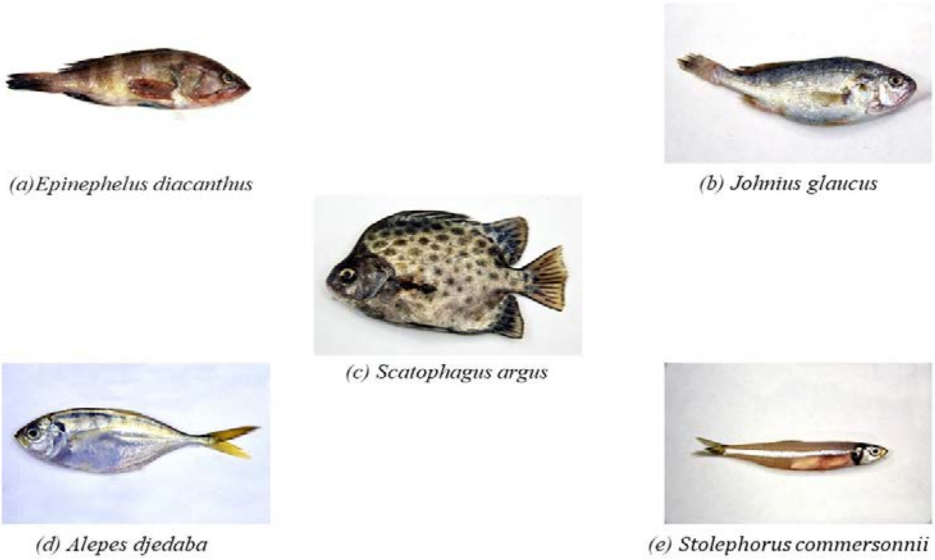


Fig. 1 Showing the photographs of five fishes (a) *E. diacanthus, (b) J. glaucus, (c) S. argus, (d) A. djedaba, (e) S. commersonnii*

2. Materials and Methods

The sample was collected from 09^{0}54.16$'$N 076^{0}05.21$'$E using HDPE Shrimp Trawl 41.6 m net. The samples were cut and flesh was taken and minced. The moisture content of the sample was determined using the gravimetric method [8]. This involved weighing a Petri dish before and after adding the sample, then drying it in an oven at 105 °C. After cooling, the dish was weighed again to determine the moisture content. The protein content was estimated using the Kjeldahl method [8]. For this, the sample was digested with concentrated sulfuric acid and a digestion mixture until the solution became colorless. The ammonia released during this process was then distilled into a boric acid solution and titrated with N/100 sulfuric acid. To estimate the fat content, the Soxhlet method [8] was employed. The sample was placed in a thimble, which was sealed with a cotton plug. This thimble was inserted into the Soxhlet apparatus, where fat was extracted by continuously washing with petroleum spirit. The extracted fat was collected in a flat-bottom flask. The ash content was determined by heating a crucible in a muffle furnace at 600 °C. After heating, the crucible was cooled in a desiccator and weighed. Then, the dried sample was added to the crucible. The sample was charred with a low flame and subsequently heated in the furnace for 3-4 hrs to obtain white or gray ash. After cooling, the crucible was weighed again, reheated for an additional 30 min, cooled once more, and weighed again to ensure a consistent result [8].

The fatty acid content was determined using the Folch method [9] for lipid extraction, which involves using chloroform-methanol mixture in a 2:1 ratio. The prepared samples were analyzed using gas chromatograph (TRACE 1300) single quadrupole mass spectrometer (1SQ 7000) equipped with a polar fused silica capillary column (TG-POLAR) with column size of 105 x 0.25mm x 0.20µm and an AI 1310 series autosampler. The mobile phase consisted of inert gas (helium), and the analysis was conducted in a high-temperature oven with the software Chromeleon 7. The temperature conditions started at 80 °C, then increased to 250 °C, with a total running time of 70 minutes. The fatty acids were separated, identified, and quantified by comparing their retention times with those obtained from a fatty acid standard.

3. Results

The protein content among the five fish species studied revealed intriguing differences. *S. argus*, known for its robust nutritional profile, topped the chart with a remarkable protein content of 14.58% (Table1). In contrast, *E. diacanthus* lagged with a modest 12.37%, highlighting the diversity in protein richness across these species. Examining the fat content, *S. argus* again emerged as a champion, boasting a fat level of 3.61%. On the other end of the spectrum, *S. commersonnii* exhibited the lowest fat content at just 1.32%, thus offering a lean option for health-conscious consumers. The analysis of ash content revealed that *A. djedaba* had the highest level at 5.27%, while *S. commersonnii* again had the least at 4.14%. Additionally, *S. commersonnii* demonstrated a remarkable moisture content of 75.59%, making it particularly appealing for culinary applications. Meanwhile, *A. djedaba* presented the lowest moisture content, although specific figures were not detailed in this study.

Table 1. Proximate composition of Five Fishes

Name of the Fish	Moisture (%)	Crude Protein (%)	Crude Fat (%)	Ash (%)
Epinephelus diacanthus	75.34	12.37	2.51	4.26
Johnius glaucus	74.14	13.20	2.37	5.14
Scatophagus argus	72.25	14.58	3.61	4.96
Alepes djedaba	69.79	14.23	1.94	5.27
Stolephorus commersonnii	75.59	13.58	1.32	4.14

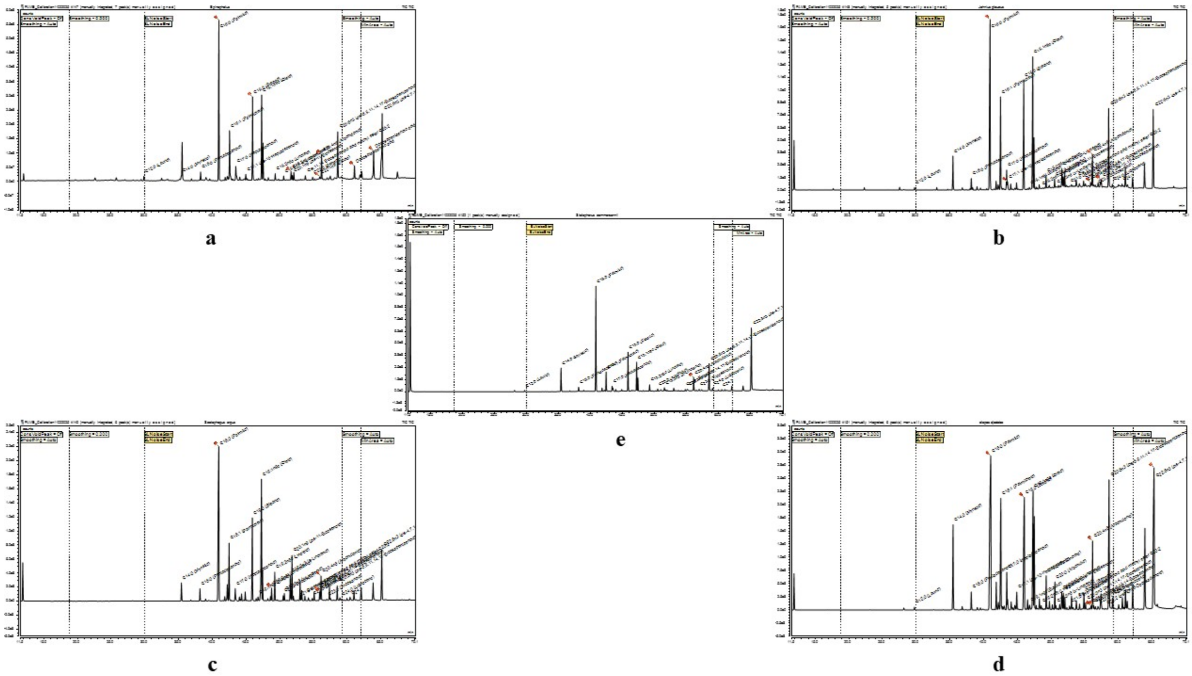
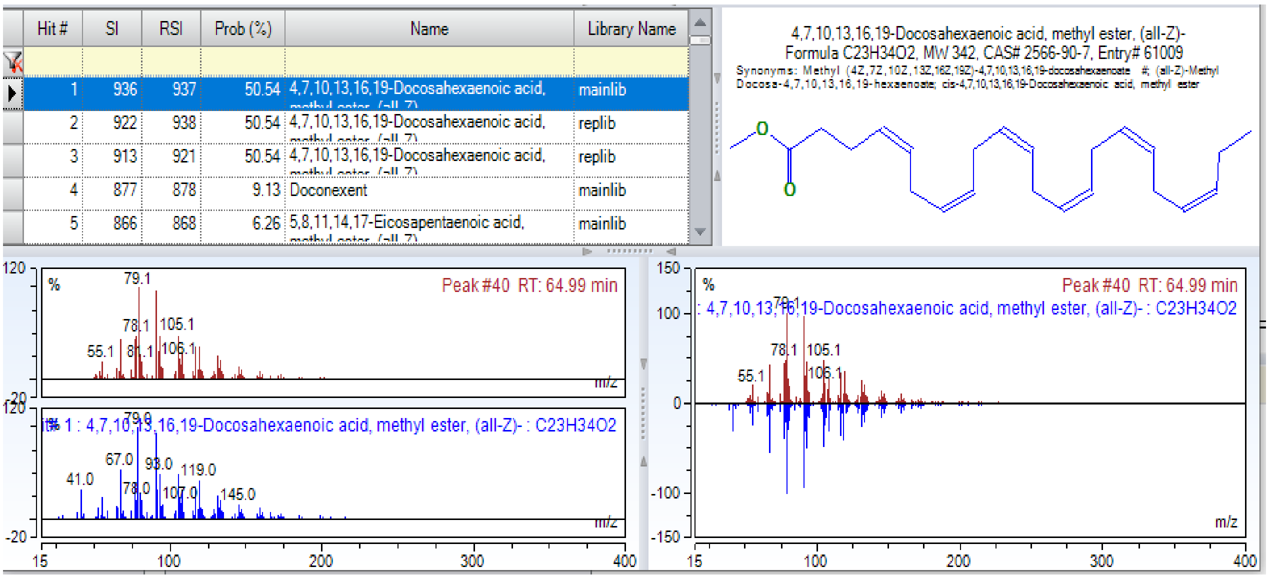


Fig. 2 The chromatogram of five fishes (a) *E. diacanthus*, (b) *J. glaucus*, (c) *S. argus*, (d) *A. djedaba*, (e) *S. commersonnii*

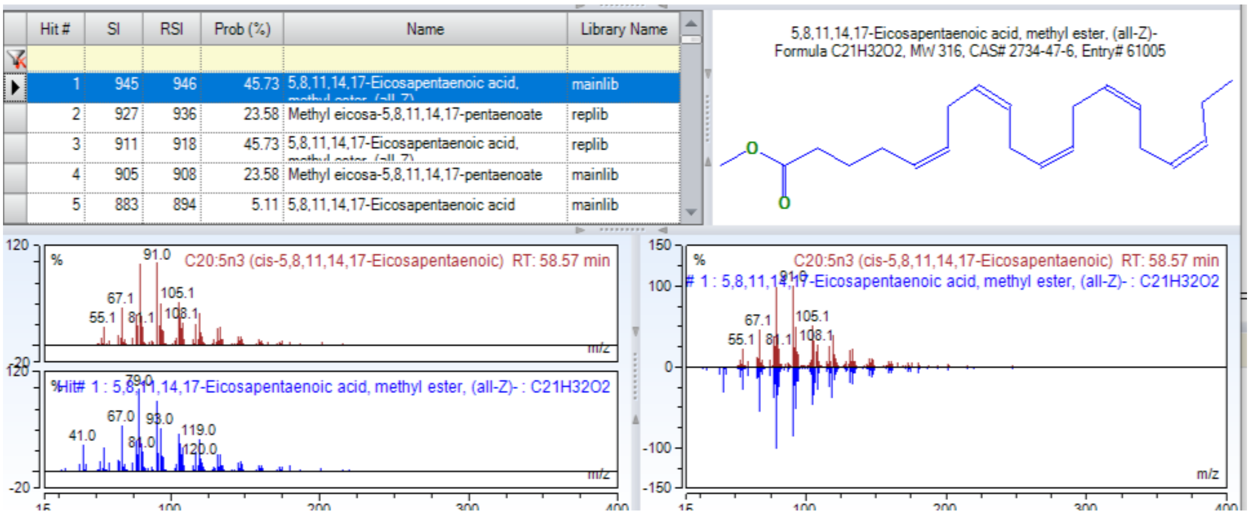
The fatty acid profiling of various fish species reveals intriguing insights into their nutritional composition, particularly concerning essential fatty acids that are vital for human health. Figure 2 illustrates the gas chromatographic profiles of five fish species—*E. diacanthus* (Fig. 2a), *J. glaucus* (Fig. 2b), *S. argus* (Fig. 2c), *A. djedaba* (Fig. 2d), and *S. commersonnii* (Fig. 2e)—while Figure 3 shows the GC-MS-based identification and mass spectral characterization of docosahexaenoic acid (Fig. 3a) and eicosapentaenoic acid methyl ester (Fig. 3b).

Among the species examined, *A. djedaba* commonly known as the shrimp scad, proudly boasts the highest concentration of eicosapentaenoic acid (EPA), attaining an impressive 9.69% (Table 2). In contrast, S. argus shows only 1.62% of EPA, highlighting the varied fatty acid profiles present among these aquatic species.

Examining docosahexaenoic acid (DHA), *Stolephorus commersonnii*, or the commerson’s anchovy, takes center stage with a remarkable DHA percentage of 23.25%. Conversely, *Scatophagus argus* trails behind with a modest DHA concentration of 7.89%. The remaining species present a varied array of DHA levels: *Epinephelus diacanthus*, a lesser-known fish, contributes 17.12%; *Johnius glaucus*, known as the pale spotfin croaker, shows 9.84%; while *Alepes djedaba* records a notable 16.89%, adding to the complexity of their nutritional profiles.



(a)



(b)

Fig. 3 Identification and Mass Spectral Analysis of (a)Docosahexaenoic Acid & (b) Eicosapentaenoic acid Methyl Ester via GC-MS.

Beyond these essential fatty acids, *Scatophagus argus* showcases a strikingly high percentage of palmitic acid at a substantial 31.88% and its shows highest percentage for both Linoleic (3.04%) and Linoenic acids (2.92%). However, its arachidonic acid content remains relatively low, measuring at 2.78%. In contrast, other examined fish species generally follow a consistent trend where the percentage of saturated fatty acids (SFA) surpasses that of monounsaturated fatty acids (MUFA) and polyunsaturated fatty acids (PUFA), *S. commersonnii* Shows highest percentage of Saturated fatty acids and polyunsaturated fatty acids and lowest for MUFA further highlighting the distinct nutritional characteristics of this particular fish. Among saturated fatty acids, palmitic acid showed the highest percentage; among monounsaturated fatty acids, oleic acid was dominant; and among polyunsaturated fatty acids, DHA exhibited the highest percentage. The ratio of n-3 fatty acids was higher than that of n-6 fatty acids in each fish species, with the highest n-3/n-6 ratio observed in *S. commersonnii.*

4. Discussion

This study investigated the nutritional value of five edible fishes caught from Kerala coast. Fish, in general, is celebrated as a unique dietary staple, prized not only for its low-fat content but also for being a concentrated source of high-quality protein. *Scatophagus argus* shows comparatively highest amount of protein. This feature makes developing innovative fish products both economically viable and nutritionally beneficial, especially as more people look for diets that are low in fat but high in healthy unsaturated fatty acids [10]. Fish and seafood products are notably nutritious, providing high levels of protein, healthy fats, and essential micronutrients [11]. Furthermore, fish is an exceptional source of high-quality, balanced, and easily digestible protein, enriched with vitamins and polyunsaturated fatty acids [12]. By increasing the availability of fish protein, we can significantly contribute to alleviating protein malnutrition [13]. In tropical regions, fish are often more accessible and affordable than other sources of animal protein, making them an essential part of a healthy diet [14].

Table 2. Fatty acid content of five fishes in percentage

Carbon no.	Fatty acids	*E. diacanthus*	*J. glaucus*	*S. argus*	*A. djedaba*	*S. commersonnii*
Saturated fatty acids (SFA)						
C 12:0	Lauric	0.75	0.26	0.00	0.17	0.43
C 14:0	Myristic	1.78	3.37	2.18	5.62	6.55
C 15:0	Pentadecanoic	1.26	1.10	1.36	0.97	1.16
C 16:0	Palmitic	27.52	24.65	31.88	20.94	27.86
C 17:0	Heptadecenoic	2.70	1.97	1.85	1.99	1.46
C 18:0	Stearic	9.70	12.21	10.49	11.60	9.46
C 20:0	Arachidic	0.00	0.83	0.51	1.46	0.54
C 21:0	Henicosanoic	0.00	0.53	0.16	0.23	0.00
C 22:0	Behenic	0.63	0.86	0.25	0.84	0.49
C 23:0	Tricosanoic	0.00	0.03	0.32	0.17	0.70
C 24:0	Lignoceric	0.00	0.35	0.15	0.67	0.70
Total		**44.34**	**46.16**	**49.15**	**44.66**	**49.35**
Monounsaturated fatty acids (MUFA)						
C 16:1	Palmitoleic acid	8.97	8.55	6.29	7.32	4.92
C 17:1	Cis-10-heptadecenoic	0.96	1.05	0.00	1.12	0.00
C 18:1n9t	Elaidic	0.00	0.30	0.53	0.35	0.00
C 18:1n9c	Oleic	12.85	13.84	15.51	9.63	7.29
C 20:1n9	Eicosenoic	0.73	1.23	4.93	0.82	0.00
C 22:1n9	Eruic	0.33	0.42	0.51	0.22	0.00
C 24:1n9	Nervonic	0.00	0.54	0.23	0.69	0.38
Total		**23.84**	**25.93**	**28**	**20.15**	**12.59**
Polyunsaturated fatty acids (PUFA)						
C 18:2n6t	Linolelaidic	0.00	0.03	1.38	0.28	0.00
C 18:2n6c	Linoleic	0.97	1.12	3.04	1.79	1.59
C18 :3n6	Linolenic	0.55	0.31	0.79	0.15	0.00
C 18:3n3	a-Linolenic	1.46	2.36	2.92	1.45	1.00
C 20:3n3	eicosatrienoic	0.00	0.70	0.89	0.35	1.24
C 20:4n6	Arachidonic	3.62	3.61	2.78	3.74	3.22
C 20:3n6	Eicosatrienoic	0.00	0.00	1.07	0.27	0.00
C 22:2	Docosadienoic	0.00	0.85	0.00	0.08	0.00
C 20:5n3	eicosapentaenoic	7.40	7.75	1.62	9.69	7.76
C 22:6n3	Docosahexaenoic	17.12	9.84	7.87	16.89	23.25
Total		**31.12**	**26.57**	**22.36**	**34.69**	**38.06**
n3		17.12	20.65	13.3	28.38	33.25
n6		5.14	5.07	9.06	6.23	4.81
n3/n6		3.33	4.07	1.47	4.55	6.70

This study reveals that *Stolephorus commersonnii* contains the highest percentage of docosahexaenoic acid (DHA, C22:6n-3) as well as the highest levels of polyunsaturated fatty acids. In contrast, *A. djedaba* is noted for its eicosapentaenoic acid (EPA, C20:5n-3) content. n-3 polyunsaturated fatty acids (PUFA) are crucial for supporting brain and visual health, and their consumption has been linked to a reduced risk of conditions such as Alzheimer's disease, dementia, and depression [15]. The remarkable levels of polyunsaturated fatty acids present in these fish species underscore their exceptional nutritional benefits. The arachidonic acid (C20:4n-6) content was highest in *Johnius glaucus*. Notably, the inclusion of essential fatty acids such as EPA (eicosapentaenoic acid), DHA (docosahexaenoic acid), and arachidonic acid significantly elevates their nutritional profile. EPA and DHA, in particular, are well-recognized for their impressive cardioprotective properties, which play a crucial role in safeguarding cardiovascular health [16]. These essential fatty acids play a crucial role in promoting overall health by reducing the risks of strokes, cognitive decline, major depressive disorders, and brain damage [17]. DHA in combination with EPA is prescribed to help prevent and treat conditions like heart disease, asthma, cancer, lung disorders, lupus, high cholesterol, high blood pressure, psoriasis, rheumatoid arthritis, bipolar disorder, digestive inflammations like ulcerative colitis, and migraines [18]. Incorporating them into the diet is crucial for enhancing overall well-being.

Linoleic acid (C18:2n-6c) and alpha-linolenic acid (C18:3n-3) were present in all the fish studied, with *S. argus* containing the highest percentage of these fatty acids. These fatty acids are essential for human nutrition because the body cannot synthesize them, yet they are necessary for tissue development [19]. In saturated fatty acids palmitic acid (C16:0) was the most abundant fatty acid followed by stearic acid (C18:0) as shown in study of marine fishes [20]. Palmitic acid (PA), the predominant saturated fatty acid in human physiology and nutrition, is integral to membrane biogenesis and functions as a crucial regulator of energy metabolism, lipid homeostasis, and diverse cellular processes [21]. In monounsaturated fatty acids oleic acid (C18:1n9c) is dominant and in polyunsaturated fatty acids DHA (C22:3n-3) is showing the highest percentage. The high concentration of beneficial polyunsaturated fatty acids in fish underscores their valuable nutritional benefits, suggesting that incorporating them into a balanced diet can contribute positively to human nutrition.

5. Conclusion

The differences in the proximate composition and fatty acid content among various fish species highlight the unique nutritional benefits each type offers to consumers. For example, *S. commersonnii* is highly valued for its significant amounts of polyunsaturated fatty acids, particularly docosahexaenoic acid (DHA), which is associated with numerous health benefits. Given the well-documented advantages of n-3 fatty acids, these fish species have the potential to greatly enhance dietary nutrition, especially in regions where fish is a primary source of protein. This study emphasizes the complex nutritional profiles of these fish, showcasing their promising role in health-oriented diets and as functional foods. However, further research is necessary to explore the bioavailability and health effects of n-3 fatty acids, ensuring a thorough understanding of their contributions to overall well-being.

6. Acknowledgement

The authors would like express sincere gratitude towards Director, ICAR-Central Institute of Fisheries Technology, Kochi, Kerala, India for providing the facilities to conduct the study and giving permission for publishing the date acquired during the study.

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