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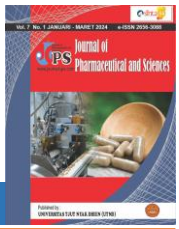

Analysis of Albumin Levels and Protein Profile of Toman Fish (*Channa Micropeltes*) from South Kalimantan

Analisis Kadar Albumin dan Profil Protein Ikan Toman *(Channa Micropeltes)* dari Kalimantan Selatan

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Abstract

Toman fish (Channa micropeltes) is a freshwater fish widely found in Kalimantan and known for its high nutritional value, particularly its albumin and omega-3 fatty acid content. However, studies regarding the metabolite profile and albumin levels of this species are still limited compared with other snakehead fish species. This study aimed to analyze the protein profile and determine the albumin levels of toman fish collected from three regions in South Kalimantan, namely Barito Kuala, Banjarmasin, and Banjarbaru. Albumin extraction was carried out using the Ultrasonic-Assisted Extraction (UAE) method. Qualitative characterization of protein functional groups was performed using ATR-FTIR spectroscopy in the range of 4000–400 cm^{-1}, while quantitative determination of albumin levels was conducted using UV–Vis spectrophotometry at a wavelength of 642 nm with bromocresol green (BCG) reagent. The results showed that the highest albumin level was obtained from Banjarmasin samples (0.560 ± 0.0343 %w/w), followed by Barito Kuala (0.542 ± 0.0416 %w/w), whereas the lowest level was observed in Banjarbaru samples (0.431 ± 0.0261 %w/w). ATR-FTIR analysis confirmed the presence of albumin in all samples through the identification of characteristic amide I (~1650 cm^{-1}) and amide II (~1550 cm^{-1}) absorption bands. Statistical analysis indicated that differences in albumin levels among sampling locations were not statistically significant ($p > 0.05$), although numerical variations were observed and may be associated with environmental and habitat conditions. Overall, the combination of UV–Vis spectrophotometry and ATR-FTIR proved effective for the characterization and determination of albumin in Channa micropeltes

***Keywords*: Albumin, ATR-FTIR, Channa micropeltes, Snakehead Fish, UV-Vis Spectrophotometry.**

Abstrak

Ikan toman (Channa micropeltes) merupakan ikan air tawar yang banyak ditemukan di Kalimantan dan dikenal memiliki nilai gizi tinggi, terutama kandungan albumin dan asam lemak omega-3. Namun, penelitian mengenai profil metabolit dan kadar albumin pada spesies ini masih terbatas dibandingkan dengan spesies ikan gabus lainnya. Penelitian ini bertujuan untuk menganalisis profil protein serta menentukan kadar albumin ikan toman yang berasal dari tiga wilayah di Kalimantan Selatan, yaitu Barito Kuala, Banjarmasin, dan Banjarbaru. Ekstraksi albumin dilakukan menggunakan metode Ultrasonic-Assisted Extraction (UAE). Karakterisasi gugus fungsi protein dilakukan menggunakan spektroskopi ATR-FTIR pada rentang bilangan gelombang 4000–400 cm^{-1}, sedangkan penetapan kadar albumin secara kuantitatif dilakukan menggunakan spektrofotometri UV–Vis pada panjang gelombang 642 nm dengan pereaksi bromkresol hijau (BCG). Hasil penelitian menunjukkan bahwa kadar albumin tertinggi diperoleh pada sampel dari Banjarmasin (0,560 ± 0,0343 %b/b), diikuti Barito Kuala (0,542 ± 0,0416 %b/b), sedangkan kadar terendah terdapat pada sampel dari Banjarbaru (0,431 ± 0,0261 %b/b). Analisis ATR-FTIR mengonfirmasi keberadaan albumin pada seluruh sampel melalui identifikasi pita serapan amida I (~1650 cm^{-1}) dan amida II (~1550 cm^{-1}). Hasil analisis statistik menunjukkan bahwa perbedaan kadar albumin antar wilayah tidak berbeda signifikan secara statistik ($p > 0,05$), meskipun terdapat variasi numerik yang diduga dipengaruhi oleh kondisi lingkungan dan habitat ikan. Secara keseluruhan, kombinasi metode spektrofotometri UV–Vis dan ATR-FTIR terbukti efektif untuk karakterisasi dan penetapan kadar albumin pada Channa micropeltes.

Kata Kunci: Albumin, ATR-FTIR, Channa micropeltes, Ikan Toman, Spektrofotometri UV-Vis.

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Introduction

Kalimantan is one of the largest islands in Indonesia and even in the world. Its rugged geographical conditions have resulted in Kalimantan having many rivers. The large number of freshwater fish species found in these rivers makes freshwater fish one of the most sought-after commodities. One type of freshwater fish that has the potential to be processed into products is the toman fish (*Channa micropeltes*). The toman fish is a carnivorous species with relatively high economic value. It has a high oil content, and toman fish oil has a higher concentration of omega-3 fatty acids than other whole fish, such as tilapia and catfish, making it a suitable choice, especially for individuals who have difficulty eating fish or require a high intake of omega-3 fatty acids.

Albumin is the main plasma protein that plays an important role in maintaining oncotic pressure, transporting endogenous and exogenous molecules, and supporting tissue repair processes [1,2]. With these vital functions, the availability of albumin has strategic value, both for health services and for the independence of national pharmaceutical raw materials. Currently, most studies in Indonesia have focused on snakehead fish (*Channa striata*), which has been proven to have high albumin levels and potential for use in the pharmaceutical field. However, toman fish (*Channa micropeltes*), which is widely distributed in Kalimantan waters, has been minimally studied in a standardized manner, particularly in relation to its albumin levels and metabolite profile [3].

Biologically and ecologically, the albumin levels and metabolite profiles of toman fish are likely influenced by geographical factors (between provinces/locations), habitat type (river, swamp, or lake), environmental parameters such as temperature, pH, dissolved oxygen, conductivity, and turbidity, as well as biometric factors such as body length and weight [4,5]. Biochemical protein theory suggests that environmental conditions can affect the secondary-tertiary structure of proteins, including albumin, through changes in amide I–II conformation that can be detected by FTIR [6]. Thus, habitat differences have the potential to produce significant variations in protein structure and related metabolites.

Methods

Place and Time of Research

The research was conducted from September 2025 to February 2026 at the Laboratory and Instrumentation of the Faculty of Pharmacy, Muhammadiyah University Banjarmasin.

Equipment

ATR-FTIR Spectrophotometer (Bruker), UV–Vis Spectrophotometer (Shimadzu), centrifuge, sonicator, centrifuge tubes, Erlenmeyer flasks, beakers, volumetric flasks, graduated cylinders, stirring rods, volumetric pipettes, propipettes, micropipettes, and quartz cuvettes.

Materials

Standard Bovine Serum Albumin (BSA), bromocresol green (BCG), acetate buffer (pH 4.2), diethyl ether, distilled water (aquadest), and 25% sodium sulfate. All chemicals used are of analytical grade purity.

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Samples

Toman fish (*Channa micropeltes*). Samples were collected from three different locations in South Kalimantan, namely Barito Kuala, Banjarmasin, and Banjarbaru. Three fish were collected from each location. The part used or collected was the meat of the Toman fish.

Procedure

Extraction and Purification of Albumin

A total of 10 g of minced toman fish flesh was mixed with 25 mL of acetate buffer solution (pH 4.2) to maintain protein stability during the isolation process. The use of acetate buffer at pH 4.2 was based on conditions below the isoelectric point of albumin (pI ±4.7), ensuring that the protein remains charged and stable in solution. The fish tissue homogenate was then extracted using the Ultrasonic-Assisted Extraction (UAE) method for 10 minutes to disrupt tissue structure and enhance protein release. After extraction, the sample was centrifuged at 5000 rpm for 30 minutes to separate the aqueous and solid phases. The resulting liquid phase was transferred into a new test tube and filtered using filter paper. Subsequently, 2 mL of 25% sodium sulfate and 2 mL of ether were added to the solution, forming a defatted aqueous phase. The mixture was then centrifuged again to obtain the supernatant containing albumin. This process resulted in two layers: an upper ether layer and a lower layer containing albumin, which was ready for analysis.

The combination of UAE, centrifugation, and defatting methods was adapted from standard protein extraction techniques from animal sources, which have been proven effective in increasing protein yield and purity [4]. The UAE method was selected because it enhances extraction efficiency through the phenomenon of acoustic cavitation, which disrupts cell walls and accelerates protein release from biological tissues. Compared to conventional methods such as maceration, UAE offers several advantages, including shorter extraction time, reduced solvent usage, higher efficiency in protein isolation, and improved extraction yield [7,8]. The extraction parameters were based on previous studies indicating that a 10-minute sonication at room temperature is effective in achieving optimal protein yield without causing significant denaturation.

Additionally, the use of ultrasound under controlled conditions has been reported to increase protein yield without causing significant denaturation [9,10]. The structural stability of the extracted protein can also be confirmed through FTIR analysis, particularly in the amide I and II bands [6].

Albumin Profiling Using IR Spectroscopy

The extracted albumin solution was placed in an ATR (Attenuated Total Reflectance) cell, then the spectrum was recorded in the wavelength range of 4000–400 cm^{-1}. The observation focused on the amide I (~1650 cm^{-1}) and amide II (~1550 cm^{-1}) bands, which are the main markers of protein secondary structure. The measured spectrum was compared with the reference albumin spectrum to confirm the presence and integrity of the protein structure in the extract [11]

Determination of Albumin Levels Using UV–Vis Spectrophotometry

Albumin levels were determined using UV–Vis spectrophotometry with bromocresol green (BCG) as the reagent at a maximum wavelength of 642 nm. The sample filtrate was mixed with the BCG reagent, incubated for 10 minutes, and then its absorbance was measured [12]. Albumin concentration was calculated based on a BSA standard calibration curve within the concentration range of 100–500 ppm [4].

This analytical method was validated through linearity, accuracy, precision, and determination of the limit of detection (LOD) and limit of quantification (LOQ) to ensure the reliability of the measurement results [13,14]. Albumin analysis requires albumin reagent in the form of bromocresol green (BCG) and a standard albumin solution prepared from Bovine Serum Albumin (BSA) [15]. The albumin content was determined by measuring the absorbance of the sample. A total of 1.5 mL of the filtered extract was mixed with 2.5 mL of 0.01% BCG reagent and allowed to stand for 10–15 minutes. The mixture was then transferred into a cuvette, and its absorbance was measured at a wavelength of 642 nm.

Data Analysis

IR spectral data were analyzed qualitatively to identify characteristic functional groups of proteins. Data obtained from UV–Vis spectrophotometric measurements were analyzed descriptively and comparatively to evaluate differences in albumin levels among sampling locations. All measurements were performed in triplicate, and the results were expressed as mean ± standard deviation (SD).

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Furthermore, to determine differences in albumin levels among the three locations (Barito Kuala, Banjarmasin, and Banjarbaru), inferential statistical analysis was conducted (e.g., one-way ANOVA followed by a post-hoc test) to confirm whether the differences were statistically significant ($p < 0.05$).

Results and Discussion

Characteristics of Toman Fish

The morphology of toman fish is characterized by a large head and wide mouth, equipped with sharp and pointed teeth used for tearing and shredding prey. Toman fish have an elongated, cylindrical, and slender body with smooth, shiny scales. Adult fish are typically bluish-black in color, with a white or pale underside. Juveniles are reddish in color, with black and orange stripes along the sides of their bodies. Along the body, there are black spots and a lateral line extending from the head to the tail. The upper part of the mouth to the caudal fin is orange, with irregular broken black boundary lines [16].

Adult toman fish can grow up to approximately 1.5 meters in length and weigh more than 10 kilograms. Toman are classified as predatory fish, feeding on various types of fish and animals such as insects and frogs. According to research by Ansyari and Slamat [17]. toman fish spawn partially throughout the year. This is supported by Sonnaria [18], who reported that members of the Channidae family are capable of spawning year-round.

Figure 1. Length of toman fish

Table 1. Length of Toman Fish (*Channa micropeltes*) from three districts in South Kalimantan

District	Sample	Length of Toman Fish (cm)	Average ± SD
Barito Kuala	1	38	38,33 ± 0,58
	2	38	
	3	39	
Banjarmasin	1	46	41,67 ± 3,79
	2	40	
	3	39	
Banjarbaru	1	41	41,00 ± 2,00
	2	43	
	3	39	

Based on **Table 1**, the average length of toman fish varies across districts. Samples from Banjarmasin showed the largest size (41.67 ± 3.79 cm), while the smallest were from Barito Kuala (38.33 ± 0.58 cm). This variation in size is influenced by environmental factors and feed availability, which are known to play important roles in the growth and biochemical composition of freshwater fish [4]. Differences in fish length may serve as an initial indicator of variations in physiological condition and protein content across regions. In addition, the proportion of body parts, from the head to the viscera, also increases accordingly [19].

To examine the relationship between biometric parameters and albumin levels, a Pearson correlation analysis was conducted between fish length and albumin concentration. The results indicated a tendency toward a positive correlation, suggesting that an increase in fish body size is associated with higher albumin levels. However, the correlation was not statistically significant ($p > 0.05$), indicating that environmental factors may have a more dominant influence than biometric factors in determining albumin levels in toman

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fish. Further studies are needed, particularly focusing on water quality parameters at the sampling sites, to better understand the factors affecting albumin levels.

This study has several limitations that should be considered. The number of samples per location (n = 3) provides only a preliminary overview and does not fully represent population heterogeneity. Additionally, environmental parameters were not measured simultaneously, and therefore could not be analyzed as covariates. Consequently, the findings of this study are exploratory in nature and require further validation through studies with larger sample sizes and integrated environmental parameter analysis.

Albumin Profiling with IR Spectroscopy

The results of ATR-FTIR analysis of the albumin filtrate showed a spectrum pattern consistent with the 1000 ppm standard albumin.

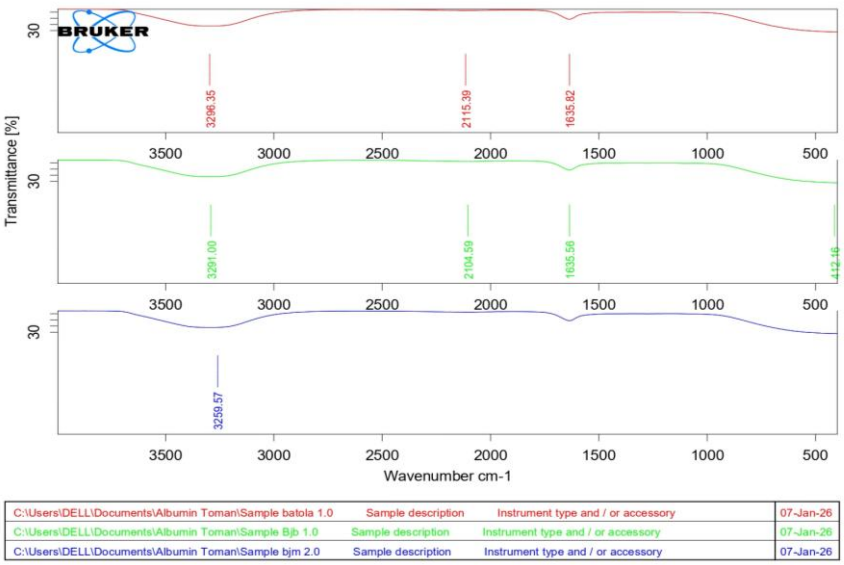

Figure 2. ATR-FTIR spectra of albumin samples from Toman fish from three districts in South Kalimantan

These results indicate that all toman fish samples contain albumin protein, although there are minor variations in chemical composition between locations. Chasanah [20] explained that high albumin content is influenced by stress levels and the environmental conditions of the habitat. Factors such as water quality, stress levels, nutrient availability, and habitat conditions can affect the stability of protein structure, which is then reflected in the FTIR spectral profile [6]. Variations in fish protein content are also thought to be influenced by differences in water quality, natural feed availability, and ecosystem factors that affect metabolism and protein synthesis [21].

ATR-FTIR spectral analysis showed that all samples exhibited characteristic protein bands, namely the amide I band around 1650 cm^{-1} and the amide II band at approximately 1540–1560 cm^{-1}. Although the overall spectral patterns were similar, minor differences were observed in band intensity and slight shifts in peak positions among the districts.

Samples from Banjarmasin exhibited relatively higher intensities in both the amide I and amide II bands compared to other locations, indicating a higher concentration of protein or albumin. This finding is consistent with the quantitative UV–Vis analysis, which showed the highest albumin levels in samples from this location. Samples from Barito Kuala demonstrated stable spectral patterns with moderate intensity, suggesting relatively well-preserved protein structures without significant conformational changes. In contrast, samples from Banjarbaru showed lower band intensities and slight broadening of the amide I band, which may indicate lower protein content or minor alterations in the secondary structure of the protein.

To strengthen the spectral interpretation, a semi-quantitative analysis was conducted by comparing the relative intensity of the amide I band to that of amide II. This intensity ratio can provide insight into changes in protein secondary structure, such as the proportion of α-helix and β-sheet. Samples from Banjarmasin showed a higher intensity ratio, indicating a dominance of more stable α-helix structures, which aligns with the higher albumin levels observed.

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Meanwhile, the broadening of the amide I band in samples from Banjarbaru suggests the possibility of conformational changes in the protein due to environmental factors. These changes may be associated with habitat conditions such as water quality and biological stress, which can influence the stability of protein structure.

Differences in the intensity and shape of the amide bands are associated with variations in protein composition as well as possible conformational changes in secondary structures, such as α-helix and β-sheet, due to environmental influences.

Overall, the ATR-FTIR results confirm that albumin is present in all samples, although with variations in structure and concentration that are consistent with differences in albumin levels among locations.

Determination of Albumin Levels with UV-Vis Spectrophotometry

Albumin levels are determined using bromcresol green (BCG) reagent at a maximum wavelength of 642 nm.

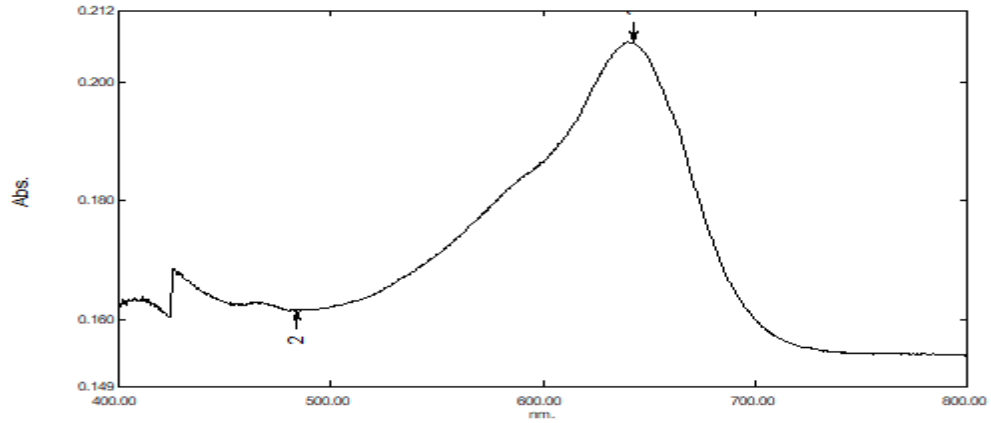

Figure 3. Absorption Spectrum of the Albumin BCG Complex at Maximum Wavelength

Based on Figure 3, the maximum absorption at 642 nm corresponds to the characteristics of the albumin-BCG complex, which is stable and sensitive to albumin concentration [12]. Measurement at this λ max results in high sensitivity to changes in protein concentration in solution.

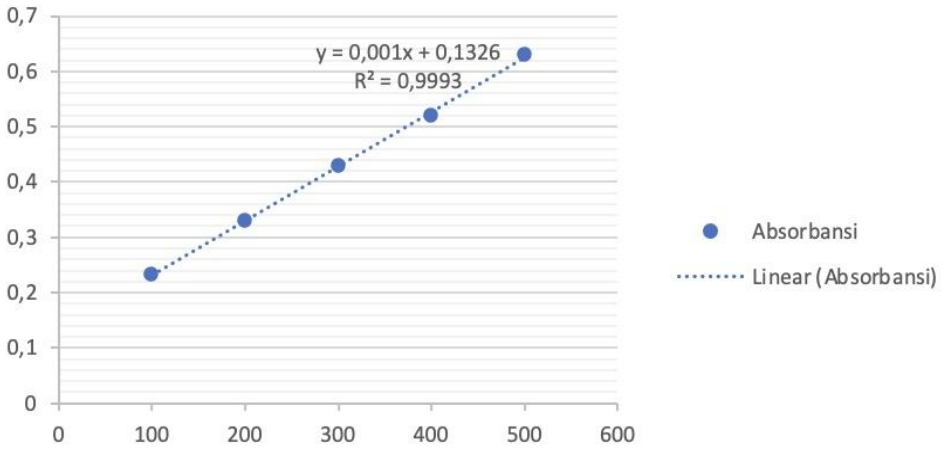

Figure 4. Standard Albumin Calibration Curve

The limit of detection (LOD) and limit of quantification (LOQ) were 23.19 ppm and 77.33 ppm, respectively, indicating that the method is sensitive enough to detect albumin at low concentrations. Accuracy testing using the spiking method yielded an average recovery of 83.31-95.59%, which is considered accurate according to the ISO/IEC standard (80-120%), while precision testing yielded an RSD of 1.40% (<2%), indicating that the method is valid and reliable [13,14]

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Variations in Albumin Levels Between Districts

Based on **Table 2,** the highest albumin content was found in the sample from Banjarmasin (0.560 ± 0.0343 %w/w), while the lowest content was obtained from Banjarbaru (0.431 ± 0.0261 %w/w). This variation indicates the influence of environmental conditions, feed availability, and physiological factors on albumin biosynthesis.

Table 2. Albumin levels in Toman fish from three districts in South Kalimantan

Sample	Level (%w/w)
Barito Kuala 1	0,5481
Barito Kuala 2	0,5796
Barito Kuala 3	0,4971
Average	0,542 ± 0,0416
Banjarmasin 1	0,5406
Banjarmasin 2	0,5991
Banjarmasin 3	0,5391
Average	0,560 ± 0,0343
Banjarbaru 1	0,4011
Banjarbaru 2	0,4401
Banjarbaru 3	0,4506
Average	0,431 ± 0,0261

The results of the analysis show that the highest average albumin content in toman fish was found in Banjarmasin, followed by Barito Kuala, and the lowest in Banjarbaru.

The highest albumin content in toman fish (*Channa micropeltes*) was found in samples from Banjarmasin. This is likely related to the more optimal aquatic environmental conditions in Banjarmasin, particularly the presence of large rivers such as the Barito and Martapura Rivers, which are nutrient-rich and have an abundant supply of natural food. In contrast, Banjarbaru has limited natural water sources, resulting in relatively lower food availability, while Barito Kuala, which is dominated by swampy areas, experiences significant fluctuations in water quality. These conditions can cause physiological stress in the fish, leading to suboptimal protein utilization for albumin synthesis [17,22,23]

Compared to previous studies, such as that reported by Pratama [4] albumin levels in toman fish show variations influenced by biometric and environmental factors. Physiologically, albumin synthesis in fish occurs in the liver and is strongly influenced by the availability of essential amino acids from natural food sources as well as environmental stress conditions. Waters with high nutrient availability tend to support optimal protein metabolism, thereby increasing albumin levels in muscle tissue. Although the Banjarmasin region faces relatively high anthropogenic pressure, the presence of major river flows with a continuous nutrient supply is believed to still support the availability of natural food for fish. This may explain the high albumin levels in samples from that region. Conversely, more limited water conditions in certain areas may affect metabolic efficiency and protein synthesis. These findings are also supported by ATR-FTIR analysis, which revealed differences in the intensity of the amide I and II bands indicators of protein secondary structure stability particularly the dominance of α-helix structures in samples with higher albumin levels.

The difference in albumin levels between districts can also be attributed to the characteristics of the habitat or structure of the fish's living environment in each region. Barito Kuala is dominated by swamp and tidal river ecosystems with relatively high water fertility and abundant natural feed availability. These conditions support optimal muscle tissue growth and protein metabolism. Banjarmasin is a riverine area influenced by urban activities and water traffic, so water quality can fluctuate but still provides sufficient food sources. Meanwhile, Banjarbaru has more stagnant waters or ponds with relatively lower natural nutrient availability compared to the main river basin.

In addition to habitat factors, the biometric size of fish also affects albumin levels. Fish with greater length and body weight generally have higher muscle mass, so total protein content, including albumin, tends to increase. This can be seen in the Banjarmasin sample, which had the highest average length and showed the highest albumin levels. Conversely, smaller body size in some samples, especially from Banjarbaru, correlates with lower albumin levels. Pratama's [4] research also reports that an increase in fish weight and length is directly proportional to an increase in protein and albumin content, due to increased protein synthesis activity during the growth phase. Thus, the variation in albumin levels in this study is influenced

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by a combination of environmental habitat factors and the biometric characteristics of fish in the form of body length and weight.

Albumin synthesis is influenced by nutrition, particularly protein intake and essential amino acids, which are the raw materials for protein synthesis in the liver, as well as physiological conditions such as hormones and disease conditions that affect hepatocyte function. Amino acids such as arginine, lysine, phenylalanine, threonine, and tryptophan are important components that support albumin synthesis [24]. The high albumin levels in fish from Barito Kuala are likely due to better water conditions and the availability of natural feed. Conversely, the low levels in Banjarmasin may be associated with high anthropogenic activity in the region. These results are in line with the research by Pratama and Ediwarman [4,20] which states that habitat variation affects functional protein levels in freshwater fish.

Overall, the combination of UV-Vis spectrophotometry and ATR-FTIR methods proved effective in determining the levels and characteristics of albumin in Toman fish. This data can serve as a basis for mapping potential raw material sources for the development of local protein-based pharmaceutical products in South Kalimantan.

Conclusions

This study demonstrated that Channa micropeltes from South Kalimantan has potential as a natural source of albumin, with the highest albumin level found in samples from Banjarmasin (0.560 ± 0.0343 %w/w) and the lowest in Banjarbaru (0.431 ± 0.0261 %w/w). ATR-FTIR analysis confirmed the presence of albumin through the characteristic amide I and amide II absorption bands, while statistical analysis showed that differences among locations were not statistically significant ($p > 0.05$). The UV–Vis spectrophotometric method using bromocresol green (BCG) also showed good validation performance with LOD and LOQ values of 23.19 ppm and 77.33 ppm, recovery of 83.31–95.59%, and RSD of 1.40%, indicating that the method is sensitive, accurate, precise, and reliable for albumin determination.

Conflict of Interest

The authors declare that there is no financial or non-financial conflict of interest related to the implementation and reporting of this research. All research processes were conducted independently without any intervention or influence from external parties.

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