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Protein Content Analysis of the Giant Mottled Eel (*Anguilla marmorata*) from Lake Talaga and Its Development as a Flipbook Learning Medium

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Abstract: This study aimed to analyze the protein content of *Anguilla marmorata* collected from Lake Talaga, Dampelas District, Donggala Regency, and to explore its potential for development as a learning medium. A descriptive quantitative approach was employed. Protein content was determined using the Biuret method coupled with UV–Vis spectrophotometry, with Bovine Serum Albumin (BSA) used as the standard. The protein contents obtained from three replicate measurements were 19.61%, 19.37%, and 20.43%, respectively, with a mean value of 19.80% and a range of 19.37–20.43%. These results indicate that the analyzed *A. marmorata* sample had a relatively high protein content, with consistent values across replicate measurements. The feasibility assessment of the developed flipbook yielded an overall mean score of 87.85%, corresponding to the "highly feasible" category. The content expert, media expert, and design expert assessments were 92.5%, 78.5%, and 88.0%, respectively, while the student response score was 92.4%. Overall, this study provides preliminary information on the protein content of *A. marmorata* from Lake Talaga and demonstrates the potential of the research findings to be integrated into a flipbook as a contextual learning resource for biology and nutrition-related topics.
Keywords: *Anguilla marmorata*; protein content; flipbook; UV–Vis spectrophotometry

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INTRODUCTION

The giant mottled eel (*Anguilla marmorata*) is a catadromous fish species widely distributed throughout Indonesian waters, including Java, Papua, Kalimantan, Sulawesi, and Sumatra (Syaifullah et al., 2023). Anguillid eels exhibit a complex migratory life cycle in which they use freshwater, brackish, and marine environments during different developmental stages, reflecting their high ecological adaptability (Arai, 2020). In Indonesia, six eel taxa have been recorded, namely *A. marmorata*, *A. celebesensis*, *A. ancestralis*, *A. borneensis*, *A. bicolor bicolor*, and *A. bicolor pacifica* (Pranata et al., 2024). This diversity highlights the ecological and fisheries importance of anguillid eels in Indonesia and their potential as biologically and economically valuable aquatic resources.

In addition to its ecological importance, *A. marmorata* has considerable nutritional potential. The species has been reported to contain relatively high levels of albumin, which is associated with physiological processes related to tissue repair and cell regeneration (Jamaluddin et al., 2020). It also contains minerals, proteins, essential fatty acids (Fitrisyah et al., 2023), and vitamin A, a micronutrient that contributes to normal immune function and visual health (Jamaluddin et al., 2025). Its protein content further supports the potential use of *A. marmorata* as an alternative source of animal protein for meeting nutritional requirements (Jamaluddin et al., 2020). However, the nutritional composition of fish may vary according to biological and environmental

conditions, including habitat characteristics, food availability, growth stage, and local ecological conditions. Consequently, nutritional data obtained from one geographical area cannot necessarily be generalized to populations from other habitats.

Central Sulawesi has considerable potential for *A. marmorata* because its landscape includes coastal waters, lakes, and river systems that can support different stages of the eel life cycle. Sulawesi is also recognized as an important region for tropical anguillid diversity. In Central Sulawesi, the presence of marine areas such as Tomini Bay together with inland freshwater habitats provides ecological connectivity that may support eel migration and population persistence (Wahid & Bestari, 2020). Nevertheless, the local utilization of *A. marmorata* as a food resource remains relatively limited. Communities around Lake Talaga generally consume fish species that are more common and readily available, whereas freshwater eels are less frequently consumed despite their occurrence in the area (Wahid & Bestari, 2020). This limited utilization indicates that the nutritional potential of local eel resources has not yet been fully recognized or incorporated into community-based food and educational contexts.

Lake Talaga, located in Dampelas District, Donggala Regency, is one of the inland habitats in Central Sulawesi where *A. marmorata* occurs. The lake supports diverse aquatic organisms and is connected to the sea through a river approximately 1 km long. Such connectivity is ecologically relevant for anguillid eels because these fishes generally grow in continental or coastal habitats and migrate toward marine spawning areas when they reach reproductive maturity (Harkai et al., 2024; Arai, 2020). Sugianti (2013) further reported that the water quality of Lake Talaga is suitable for freshwater eels, suggesting that the lake represents an important habitat for this species. Despite its ecological relevance, scientific information on the nutritional composition of *A. marmorata* from Lake Talaga remains scarce, particularly with respect to protein content.

Previous studies have examined the nutritional characteristics of *A. marmorata* from other waters in Central Sulawesi, including the Palu River and Lake Poso (Jamaluddin et al., 2019; 2020). However, comparable data from Lake Talaga have not been adequately documented. This constitutes an important local knowledge gap because differences in habitat quality, water characteristics, natural food resources, and other environmental factors may influence fish nutritional composition. Therefore, determining the protein content of *A. marmorata* collected from Lake Talaga can provide baseline information on this locally available fisheries resource. In the present study, protein was determined using the Biuret method combined with UV-Vis spectrophotometry, a colorimetric approach commonly used for quantitative protein determination based on the formation of a colored complex between peptide bonds and copper ions under alkaline conditions (Masriah et al., 2025).

The lack of locally generated biological information also has implications for biology education. Nutrition and biological-resource topics are frequently presented using generalized examples, with relatively limited integration of organisms and phenomena that students encounter in their own environments (Destiara, 2020; Nurlaeliana et al., 2022). Learning that incorporates local biological resources can provide a meaningful connection between scientific concepts and students' environmental contexts (Adriana et al., 2024). Evidence from science education research further indicates that incorporating local and indigenous knowledge can make science learning more contextual by linking formal scientific concepts with knowledge and practices embedded in students' communities (Latip et al., 2024). Similarly, the use of locally contextualized biological materials has been associated with

opportunities to strengthen students' biological literacy and their ability to connect biological knowledge with real-world situations (Hidayat et al., 2024).

Digital learning resources offer an additional means of presenting such locally contextualized scientific information. Flipbooks combine text, images, tables, and graphical representations in a structured digital format that can be accessed through computers, smartphones, and tablets (Sari & Ahmad, 2021). Previous studies have reported that digital learning media can support student engagement, self-directed learning, and active interaction with biological content (Wijayanti & Isnawati, 2023; Unayah et al., 2024). More broadly, research on digital learning in biology has shown that technology-supported resources can promote active learning and provide more interactive ways of organizing and engaging with scientific content (Metcalf et al., 2023; Bilici & Yilmaz, 2024). Therefore, transforming locally generated laboratory findings into a digital flipbook provides a means of linking empirical biological data with contextual learning materials.

The novelty of this study lies in the integration of two complementary components. First, it provides baseline information on the protein content of *A. marmorata* collected from Lake Talaga, for which location-specific nutritional data remain limited. Second, the laboratory findings are incorporated into a digital flipbook designed as a contextual biology learning resource. Unlike learning materials that rely primarily on generalized biological examples, the developed flipbook uses locally generated scientific data to connect nutrition concepts with a fisheries resource from Central Sulawesi. This approach is expected to strengthen the relevance of biology learning by linking laboratory evidence, local biodiversity, and nutrition-related concepts without separating scientific investigation from its educational application.

Based on these considerations, this study aimed to (1) determine the protein content of *A. marmorata* collected from Lake Talaga using the Biuret method with UV-Vis spectrophotometry and (2) develop the research findings into a digital flipbook and evaluate its feasibility as a contextual learning resource for nutrition-related topics in biology.

METHOD

This study employed a quantitative descriptive design to analyze the protein content of marbled eel (*Anguilla marmorata*) collected from Lake Talaga using the Biuret method coupled with UV–Vis spectrophotometry. Protein content was determined from sample absorbance values using a calibration curve constructed with Bovine Serum Albumin (BSA) as the protein standard, allowing quantitative estimation of protein concentration. The resulting data were subsequently used as the scientific basis for developing a digital flipbook as a learning medium. The feasibility of the developed flipbook was evaluated through expert validation and student responses.

The study sample consisted of one *A. marmorata* individual collected from Lake Talaga, Donggala Regency, Central Sulawesi (Figure 1). Species identification was based on morphological characteristics using an eel identification key. Diagnostic characteristics included an elongated, eel-like body; a marbled color pattern; continuous dorsal, caudal, and anal fins; and other morphological features consistent with the description of *A. marmorata*. Protein analysis was performed in three technical replicates using homogenized samples obtained from the same individual.

Figure 1. Sampling location

The primary instrument used in this study was a UV–Vis spectrophotometer for measuring solution absorbance. Supporting equipment included graduated cylinders, test tubes, measuring pipettes, test-tube racks, Erlenmeyer flasks, an analytical balance, water bath, centrifuge, shaker, volumetric flasks, micropipettes, Petri dishes, filter paper, and mixing equipment. The materials included a 5,000 ppm BSA stock solution for preparing the protein calibration standards, Biuret reagent for protein determination, 1 M NaOH as the sample extraction solution, and distilled water for standard dilution and sample preparation.

Preparation of the BSA Calibration Curve

A protein calibration curve was prepared from a 5,000 ppm BSA stock solution. Standard solutions at concentrations of 1,000, 2,000, 3,000, 4,000, and 5,000 ppm were prepared by transferring 0.2, 0.4, 0.6, 0.8, and 1.0 mL of the 5,000 ppm BSA stock solution, respectively, and adding distilled water to a final volume of 1 mL. Subsequently, 3 mL of Biuret reagent was added to each standard solution, followed by incubation for 30 min. Absorbance was measured using a UV–Vis spectrophotometer at the maximum wavelength previously determined by scanning the 5,000 ppm BSA standard solution. The resulting absorbance values were used to construct the calibration curve and obtain the linear regression equation.

Sample Preparation and Protein Analysis

For the first and second technical replicates, 5.03 g of eel muscle tissue was weighed, whereas 5.16 g was used for the third replicate. Each sample was transferred to an Erlenmeyer flask, followed by the addition of 25 mL of distilled water and 5 mL of 1 M NaOH. The mixture was heated in a water bath at approximately 90°C for 15 min and subsequently homogenized using a shaker for 2–3 h. The mixture was then centrifuged at 5,000 rpm for 45 min to obtain a clear supernatant.

A 1-mL aliquot of the resulting supernatant was mixed with 3 mL of Biuret reagent and incubated for 30 min. Absorbance was then measured using a UV–Vis spectrophotometer at the predetermined maximum wavelength. Protein content was calculated from the established BSA calibration curve using the following equation (Tellu et al., 2024):

$$\textbf{Protein Content}\ (\%) = \frac{\left(\frac{\boldsymbol{x}}{\mathbf{1000}}\right) \times \boldsymbol{Y} \times \boldsymbol{Fp}}{\boldsymbol{W}}\ \mathbf{100}\%$$

Note:
x = BSA-equivalent concentration of the sample (mg/L)
Y = extract volume (L)
W = sample weight (g)
Fp = dilution factor

Development of the Flipbook Learning Medium

The findings from the protein analysis were subsequently incorporated into a digital learning medium in the form of a flipbook (e-book). The flipbook comprised 15 pages covering the morphology and taxonomy of *A. marmorata*, the benefits of eel, the principles of protein analysis using UV–Vis spectrophotometry, the Biuret protein assay procedure, the results of the eel protein analysis, and a concluding section. The learning medium also included images illustrating eel morphology, an illustration of a UV–Vis spectrophotometer, the BSA calibration curve, a table presenting the research findings, and supporting references. These components were designed to provide a contextual learning resource for nutrition-related topics.

Validation and User-Response Assessment

The learning medium was evaluated by three expert validators: one content expert, one media expert, and one design expert, all of whom were lecturers in the Biology Education Study Program. In addition, a user-response assessment was conducted with 25 sixth-semester Biology Education students who had previously completed nutrition-related coursework.

The validation instrument employed a five-point Likert scale. Content validation assessed the appropriateness of the learning material, conceptual accuracy, clarity of presentation, and relevance of the supporting sources. Design validation covered visual appearance, layout, typography, and visualization, whereas media validation assessed the overall quality of the medium, readability, appropriateness of images, and presentation of information. The student-response assessment evaluated the readability, ease of use, and attractiveness of the learning medium.

The feasibility percentage was calculated from the validation scores using the following equation:

$$\boldsymbol{P} = \frac{\sum \mathbf{x}}{\sum \mathbf{xi}}\ \mathbf{x\ 100\%}$$

Note:
P = percentage score
Σx = mean response score
Σx_i = total score for the response categories
The resulting percentage was then interpreted according to the feasibility criteria proposed by Arikunto (2010), as presented in Table 1.

Table 1. Feasibility criteria for the flipbook learning medium

Percentage (%)	Feasibility Category
81–100	Very feasible
61–80	Feasible
41–60	Moderately feasible

Percentage (%)	Feasibility Category
21–40	Less feasible
0–20	Not feasible

RESULTS AND DISCUSSION

Protein Content of *Anguilla marmorata*

The absorbance values of the BSA standard solutions measured at a wavelength of 550 nm are presented in Table 2.

Table 2. Absorbance measurements of BSA standard solutions

BSA Concentration (ppm)	Absorbance at 550 nm
1000	0.050
2000	0.127
3000	0.202
4000	0.273
5000	0.352

As shown in Table 2, the absorbance of the BSA standard solutions increased proportionally with increasing concentration. This relationship indicates that protein concentration is directly proportional to absorbance; thus, higher protein concentrations produce higher absorbance values (Mashitoh et al., 2025). The absorbance data were subsequently used to construct a standard calibration curve. Based on the calibration curve shown in Figure 2, the linear regression equation was $y = 8 \times 10^{-5}x - 0.0242$, with an R^2 value of 0.9998. The coefficient of determination approaching 1 indicates a very strong linear relationship between BSA concentration and absorbance, supporting the use of the calibration curve for determining protein concentrations in the analyzed samples.

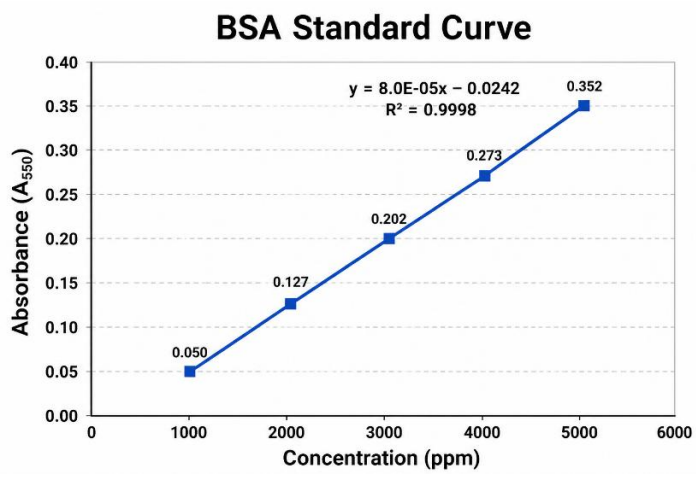

Figure 2. BSA calibration curve

The observed linear relationship is consistent with the Beer–Lambert law, which states that absorbance is directly proportional to concentration within a specified analytical range. Accordingly, an increase in concentration results in a proportional increase in absorbance during UV-Vis spectrophotometric analysis (Sulistyani et al., 2025). The results of the protein content analysis of *A. marmorata* are presented in Table 3.

Table 3. Protein content analysis of eel (*Anguilla marmorata*)

Sample	Sample Code	Absorbance at 550 nm	BSA Concentration (mg/L)	Volume (L)	Sample Weight (g)	Protein Content (%)	Mean (%)
Eel	U1	0.469	6576.000	0.03	5.03	19.61	
	U2	0.463	6496.000	0.03	5.03	19.37	**19.80**
	U3	0.503	7029.333	0.03	5.16	20.43	

The results presented in Table 3 show that the measured protein content of *A. marmorata* from Lake Talaga was relatively consistent across the three technical replicates, with an average value of 19.80%. This value corresponds to approximately 19.80 g of protein per 100 g of fresh eel flesh. The slightly higher value observed in U3 than in U1 and U2 may be associated with technical factors, including differences in sample weight, homogenization efficiency, extraction procedures, and the precision of absorbance measurements.

The mean protein content obtained in this study is comparable to values previously reported for fresh eel. Nafsiyah et al. (2018) reported protein contents of 17.30% in *A. marmorata* and 16.78% in *A. bicolor bicolor*, while a study cited by Nafsiyah et al. (2018) reported approximately 19.54% protein in fresh *A. marmorata* from Vietnam. These findings indicate that eel muscle contains a substantial amount of protein, although the absolute values may vary among species, populations, habitats, and analytical conditions.

The nutritional relevance of eel is also supported by its amino acid composition. Jamaluddin et al. (2019) reported that *A. marmorata* and *A. bicolor* contain both essential and non-essential amino acids. Therefore, the relatively high protein content observed in the present study supports the potential of *A. marmorata* as a local animal-based protein source. However, overall protein quality cannot be inferred from total protein concentration alone and should ideally be evaluated together with amino acid composition, digestibility, and bioavailability (Mohanty et al., 2014).

The protein content measured in the present study can also be compared with several commonly consumed freshwater fish. Previous studies reported protein contents of approximately 16.05% in Nile tilapia (*Oreochromis niloticus*), 16.00% in common carp (*Cyprinus carpio*), 14.53% in pangasius catfish (*Pangasius* sp.), and 13.18–17.70% in catfish (*Clarias* sp.) (Matondang et al., 2022; Ambarwati et al., 2025). These comparisons suggest that the protein content measured in the analyzed *A. marmorata* specimen is comparable to, and in some cases higher than, that reported for several commonly consumed freshwater fish. Nevertheless, direct interspecific comparisons should be interpreted cautiously because protein concentration may vary according to analytical method, moisture content, tissue characteristics, body size, feeding conditions, and physiological status.

Variation in fish protein content may also be influenced by developmental stage, season, feeding conditions, and environmental characteristics (Nafsiyah et al., 2018; Shearer, 1994). In anguillid eels, developmental transitions from glass eel to silver eel involve substantial physiological changes that may alter nutrient storage and body composition. Natural food availability and habitat conditions may further influence nutrient assimilation. However, these variables were not directly measured in the present study and should therefore be considered as possible explanations rather than confirmed determinants of the observed protein content.

An important analytical limitation should be considered when interpreting the results. The absorbance values of the samples (0.463–0.503) were higher than the maximum absorbance of the BSA standards (0.352), indicating that the calculated

sample concentrations were obtained by extrapolation beyond the established calibration range. Although the calibration curve showed excellent linearity (R^2 = 0.9998), future analyses should preferably dilute the samples so that their absorbance values fall within the validated calibration range or extend the calibration range using additional standards.

Another limitation is that the three measurements were technical replicates derived from a homogenized sample of a single eel individual. Therefore, the small variation among U1, U2, and U3 reflects analytical repeatability rather than biological variation within the Lake Talaga population. Consequently, the mean protein value of 19.80% should be interpreted as the protein content of the specimen analyzed in this study and should not yet be generalized to the entire *A. marmorata* population of Lake Talaga. Future studies should include multiple independent individuals representing different body sizes, developmental stages, seasons, and sampling locations.

Despite these limitations, the present study provides preliminary information on the protein content of *A. marmorata* from Lake Talaga. These findings may serve as baseline data for further nutritional and ecological investigations. Broader biological sampling and complementary analyses of amino acids, lipids, micronutrients, and food safety would strengthen the assessment of the species as a potentially valuable local food resource.

Feasibility of the Flipbook as a Learning Medium

Learning media are instructional tools used to present educational content through visual or digital formats, thereby attracting learners' attention and facilitating their understanding of the concepts being taught (Masduki et al., 2024). In this study, a digital flipbook was developed based on the results of the protein analysis of *A. marmorata*. The feasibility of the learning medium was assessed by a content expert, a media expert, a design expert, and a group of 25 students. The feasibility assessment results are presented in Table 4.

Table 4. Feasibility assessment of the learning medium

No.	Evaluator	Validation Score (%)	Category
1	Content expert	92.5	Highly feasible
2	Media expert	78.5	Feasible
3	Design expert	88.0	Highly feasible
4	Student group (n = 25)	92.4	Highly feasible
	Mean	**87.85**	**Highly feasible**

As shown in Table 4, the flipbook achieved an average feasibility score of 87.85%, which falls within the highly feasible category according to Arikunto (2010). The content expert provided the highest validation score (92.5%), followed by the student group (92.4%) and the design expert (88.0%). The media expert assigned a score of 78.5%, indicating that the learning medium was feasible but that certain technical aspects could still be improved.

The high content-validation score indicates that the laboratory findings on *A. marmorata* were appropriately transformed into instructional material. Integrating original research data into the flipbook provides a contextual learning approach because students are introduced not only to general concepts of protein and nutrition but also to empirical information derived from a locally relevant biological resource.

The digital format of the flipbook also enables the integration of text, images, tables, graphs, and laboratory procedures within a single learning resource. Such multimodal presentation may help organize scientific information more systematically and facilitate students' understanding of relationships among biological concepts, numerical data, and experimental procedures. Previous studies have similarly reported positive feasibility and student responses toward digital flipbooks in biology and science education (Wijayanti & Isnawati, 2023; Unayah et al., 2024).

However, the relatively lower score provided by the media expert suggests that some technical components still require refinement. Improvements may be needed in navigation, visual consistency, typography, image quality, readability, and interactive features. Addressing these aspects may enhance the usability of the flipbook and ensure that the visual elements effectively support the scientific content.

The student response score of 92.4% indicates strong acceptance of the developed medium. Nevertheless, a positive user response should not be interpreted as direct evidence of instructional effectiveness because the present study assessed feasibility and user perceptions rather than measurable learning outcomes. No pre-test–post-test design, control group, or experimental comparison was conducted. Future studies should therefore evaluate whether the flipbook improves conceptual understanding, learning motivation, scientific literacy, or academic achievement compared with conventional learning materials.

Overall, the main educational contribution of the developed flipbook lies in its integration of local biological resources, laboratory-generated nutritional data, and digital learning media. This approach has the potential to make biology learning more contextual and relevant by linking concepts of nutrition, biodiversity, laboratory analysis, and local fishery resources.

CONCLUSION

Based on the findings of this study, the marbled eel (*Anguilla marmorata*) collected from Lake Talaga, Donggala Regency, had an average total protein content of 19.80%, as determined using the Biuret method with UV–Vis spectrophotometry. These findings indicate that *A. marmorata* has a relatively high protein content and may serve as a valuable source of local biological information regarding the nutritional characteristics of eel. The flipbook-based learning media developed from the research findings achieved an average feasibility score of 87.85%, which was classified as highly feasible based on expert validation and student responses. These results demonstrate that the developed flipbook has the potential to serve as a contextual learning resource for supporting biology instruction, particularly on nutrition-related topics.

RECOMMENDATION

Future studies should expand the assessment of protein content in *A. marmorata* by including samples from multiple aquatic habitats to investigate potential variations associated with differences in environmental conditions. In addition, alternative analytical methods, such as the Kjeldahl or Bradford method, should be considered for comparison to provide more comprehensive results and strengthen data validity. Further development of the flipbook-based learning media may also incorporate interactive multimedia features, such as animated videos, to enhance learner engagement and improve its educational effectiveness.

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