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Analysis of Total Protein and Non-Protein Nitrogen in Durian Seeds and Durian Flour Using the Kjeldahl Method

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Abstract: Durian seeds, often regarded as agricultural waste, contain notable amounts of macronutrients, particularly proteins, that warrant further exploration for potential food and feed applications. This study aimed to evaluate the total nitrogen, protein, and non-protein nitrogen (NPN) contents in fresh and processed seeds of three local durian (Durio zibethinus) cultivars Bintana, Pelangi, and Bawor using the Kjeldahl method. Fresh seeds and their corresponding flours were analyzed for moisture content, total nitrogen, and protein fractions, including actual protein and NPN levels. The results revealed significant variations across cultivars and between fresh and processed samples. Bintana exhibited the highest protein content in both fresh and flour forms, followed by Pelangi and Bawor. Processing, including soaking, drying, and flour preparation, was found to increase measurable protein content but may also contribute to protein denaturation. Overall, durian seeds, particularly from the Bintana variety, demonstrate promising nutritional potential as a functional food ingredient or alternative protein source.

Keywords: Durian seed; Food waste utilization; Kjeldahl method; Non-protein nitrogen; Protein analysis.

Introduction

Proteins are vital macromolecules that play a fundamental role in the structure and function of all living organisms (Juan et al., 2021; Morris et al., 2022). They are composed of long chains of amino acids linked by peptide bonds, forming complex polypeptides. These macromolecules make up approximately 50% of the dry weight of living cells and perform various biological functions including enzymatic activity, structural support, and nutrient transport. Proteins are primarily made up of the carbon (C), hydrogen (H), oxygen (O), and nitrogen (N), and may also contain sulfur (S) and phosphorus (P). When subjected to hydrolysis using acidic solutions or enzymatic action, proteins break down into their constituent amino acids (Lin et al., 2022; Soto-Sierra et al., 2018; Wang et al., 2019; Zhou & Pang, 2018). This hydrolysis process is fundamental to understanding the nutritional content of foods, especially in determining the bioavailability of nitrogen and amino acids.

Durian *(Durio zibethinus)*, known as the "king of fruits," is one of the most popular tropical fruits in Southeast Asia (Husin et al., 2018; Van Hau et al., 2023). Native to Indonesia, durian is widely consumed during its peak season and is highly valued for its rich, creamy flesh and distinctive aroma (Wiangsamut & Wiangsamut, 2023). In Indonesia, the durian fruit season is a much-anticipated event, as the fruit is only available during specific times of the year and is considered a delicacy (Ali et al., 2025). While most research and commercial interest has focused on the edible flesh of the fruit, the seeds, which are typically discarded as waste, also hold considerable nutritional value.

Several studies have suggested that durian seeds contain significant amounts of macronutrients, particularly carbohydrates and proteins, making them a potential resource for food and feed applications (Baqi et

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al., 2021; Khaksar et al., 2024; Purnama et al., 2022). The seeds of durian are generally round or elongated, depending on the cultivar, and are often overlooked despite being rich in important nutrients. A chemical analysis of durian seeds has shown the presence of various amino acids, which are the building blocks of proteins. These include aspartic acid (0.87%), glutamic acid (0.97%), serine (0.33%), histidine (0.13%), glycine (0.33%), threonine (0.31%), arginine (0.42%), alanine (0.42%), tyrosine (0.34%), methionine (0.07%), valine (0.40%), phenylalanine (0.35%), isoleucine (0.34%), leucine (0.52%), and lysine (0.36%) (Amid et al., 2012; Kostryco & Chwil, 2022; Retnoningsih et al., 2016; Tongdee, 1992). These amino acids are essential for human nutrition and indicate that durian seeds could serve as a valuable protein source.

Despite their potential, the protein content of durian seeds can vary significantly due to post-harvest handling and processing methods such as soaking, drying, and exposure to heat and sunlight (Le, 2024). These processes can lead to protein denaturation, breaking of peptide and hydrophobic bonds, and the loss of nutritional quality. Additionally, a distinction must be made between true protein content and non-protein nitrogen (NPN) compounds, such as free amino acids, urea, and other nitrogen-containing compounds, which may not contribute to the nutritional value in the same way as intact proteins. This distinction is important when evaluating the protein quality and suitability of durian seeds for dietary use.

Quantifying both total protein and NPN content provides a more accurate assessment of the nutritional potential of durian seeds. The Kjeldahl method is a widely accepted and reliable technique for determining total nitrogen content in organic materials (Gunamantha et al., 2021). By using this method, it is possible to estimate both protein-bound nitrogen and non-protein nitrogen fractions, thus offering a comprehensive picture of the seed's protein profile (Hamim et al., 2025; Ismail & Smith, 2024).

In this study, we aim to evaluate the total protein and non-protein nitrogen (NPN) content in durian seeds and their corresponding flours prepared from three durian cultivars: Bintana, Pelangi, and Bawor. These cultivars, commonly found in the Dairi region of Indonesia, were selected for their availability and local importance. By applying the Kjeldahl method, this research seeks to provide baseline data on the nutritional composition of durian seeds, with a focus on their potential use as a functional ingredient in food products or as a feed source.

Method

Materials

The primary material used in this study was durian *(Durio zibethinus)* seed flour, obtained from three local cultivars—Bintana, Pelangi, and Bawor—sourced from the Dairi District, North Sumatra, Indonesia. The seeds were processed into flour before analysis. The chemicals included sulfuric acid (98%), copper sulfate ($CuSO_4$), potassium sulfate (K_2SO_4), sodium hydroxide (NaOH), trichloroacetic (TCA), methyl red, methylene blue, and phenolphthalein were purchased without further purification. The standardized solution using in this work was prepared as described by Yao et al. (2024).

NPN Determination

A 10 g sample of durian seed flour was weighed and transferred into a Kjeldahl flask. A catalyst mixture consisting of 2.5 g of potassium sulfate (K_2SO_4) and copper (II) sulfate pentahydrate ($CuSO_4 \cdot 5H_2O$) in a 1:1 ratio was added. Then, 15 mL of concentrated sulfuric acid (H_2SO_4) was introduced, and the mixture was digested for 2–3 hours until the solution turned a clear green color, indicating complete digestion.

After cooling, 100 mL of distilled water was added. Then, 10 mL of 40% sodium hydroxide (NaOH) was carefully added to alkalize the solution and facilitate ammonia liberation. The flask was connected to a distillation apparatus, and the liberated ammonia was captured in a receiving flask containing 10 mL of 0.1 N sulfuric acid and 2–3 drops of a mixed indicator solution (methyl red and methylene blue).

The distillate was collected until a volume of approximately 10 mL was obtained. Titration was then performed using standardized 0.02 N NaOH until the color changed to a stable green endpoint. These values were subsequently used to calculate total nitrogen, total protein, and non-protein nitrogen (NPN) content in the durian samples.

Determination of Water Content in Durian Seeds

The moisture content of durian seed samples was determined using a standard gravimetric method. Aluminum cups and their lids were first dried in an oven at a temperature of 100 to 102 degrees Celsius for 15 minutes, cooled in a desiccator for 20 minutes, and then weighed to record the tare weight. Approximately 5 grams of homogenized durian seed flour was placed in each cup. The samples were then dried in an oven at 100 to 102 degrees Celsius for 3 to 5 hours, ensuring that the cups did not touch the oven walls.

For samples that were resistant to decomposition by prolonged heating, overnight drying for approximately 16 hours was performed. After drying, the samples were cooled in a desiccator and weighed

again. To ensure accuracy, the drying and weighing process was repeated in 30-minute intervals until a constant weight was achieved. A stable weight was defined as a difference of less than 0.2 milligrams between two consecutive weighing. This procedure ensured reliable and reproducible moisture content determination in accordance with the method outlined in reference (Yao et al., 2024).

Determination of Total Nitrogen and Protein Content

The total nitrogen (N-total) and total protein content of the durian seed samples were determined using the Kjeldahl method. A known weight of the dry sample was transferred into a Kjeldahl digestion flask, to which 2 grams of catalyst mixture (equal parts potassium sulfate and copper sulfate) and 3 milliliters of concentrated sulfuric acid (H_2SO_4) were added. The mixture was then digested for approximately 3 hours until the solution turned a clear green, indicating complete decomposition of organic material.

After digestion, the solution was allowed to cool and then diluted with 10 milliliters of distilled water. The contents were transferred into an Erlenmeyer flask, and 15 milliliters of 40 percent sodium hydroxide (NaOH) solution were added to render the mixture strongly alkaline. This step released ammonia gas from the digested ammonium sulfate. The flask was then connected to a distillation apparatus.

Ammonia was distilled into a receiving flask containing 25 milliliters of 0.02 normal sulfuric acid (H_2SO_4) and 3 drops of a mixed methyl red–methylene blue indicator. The titration was performed using 0.02 normal sodium hydroxide (NaOH) until the color of the solution shifted from purple to green, marking the endpoint. A blank determination was also conducted using the same procedure but without the sample, to account for background nitrogen levels.

The total nitrogen content was calculated using the volume of titrant consumed, and the total protein content was obtained by multiplying the nitrogen content by the protein conversion factor of 6.25, which is standard for plant-based materials. For fresh samples, the nitrogen and protein contents were back calculated from dry basis values using the measured moisture content.

Separation of Protein from Non-Protein Nitrogen (NPN)

Protein separation from non-protein nitrogen (NPN) was achieved through precipitation using a 10% trichloroacetic acid (TCA) solution. A weighed dry sample was placed in a 200 mL beaker, and 50 mL of distilled water was added. The mixture was allowed to stand for 30 minutes to hydrate the sample. Subsequently, 10 mL of 10% TCA was added, and the solution was incubated for an additional 30 minutes to facilitate protein precipitation. Afterward, the mixture was filtered, and the resulting precipitate, which contained the protein fraction, was washed twice with the TCA solution to remove any residual non-protein nitrogen (NPN) content.

The total nitrogen content of the sample was determined using the Kjeldahl method, as outlined in standard protocols. The dry sample was placed in a Kjeldahl digestion flask, and 2 g of catalyst mixture and 3 mL of concentrated sulfuric acid (H_2SO_4) were added. The sample was digested for approximately 3 hours, until the digestate turned clear green, indicating the complete breakdown of organic matter.

After digestion, the sample was cooled, and 10 mL of distilled water was added. The solution was transferred into a 250 mL Erlenmeyer flask, followed by the addition of 15 mL of 40% sodium hydroxide (NaOH), which made the solution alkaline and caused the color to shift to black. The mixture was then subjected to distillation to release ammonia gas (NH_3).

The ammonia was captured by a standard 0.02 N hydrochloric acid (HCl) solution in the receiving flask. To improve the contact between ammonia and acid, the end of the distillation tube was immersed as deeply as possible into the HCl solution. Distillation continued until no further ammonia was detected, as indicated by the absence of alkalinity in the receiving flask (determined by the red litmus paper remaining red).

Following distillation, 25 mL of 0.02 N H_2SO_4 and 3 drops of a mixed indicator were added to the receiving flask. The distillate was titrated with 0.01 N NaOH until the color changed from purple to green, indicating the end of titration. A blank determination was performed using the same procedure but without the sample, to account for any procedural or reagent-related variations.

The total nitrogen content was calculated using Formula 1.

$$\%\ N\ (total) = \frac{v\ NaOH(blank-sample)x\ N\ NaOH\ x\ 14.008\ x\ 100\%}{mass\ x\ 100} \quad (1)$$

where, N NaOH = Normality of NaOH after standardization. The total protein content was calculated using the following expression, where the durian seed conversion factor = 6.25, while the total protein was obtained by Formula 2.

$$\text{Total Protein (\%)} = \%\ \text{N-total x conversion factor} \quad (2)$$

The NPN content was calculated as the difference between the total nitrogen content and the nitrogen content associated with the protein fraction according to Formula 3 (Harvey, 1999).

$$\text{Total content NPN from N total (\%)} = \frac{\%\ NPN\ x\ 100}{\%\ N-NPN\ total\ content} \quad (3)$$

During distillation, the results of the destruction are diluted with distilled water. This dilution is necessary to reduce the intensity of the reaction that will occur later if the solution is added with an alkali compound. The solution is made alkaline by adding sodium hydroxide. The purpose of adding sodium hydroxide is to break down the ammonium sulfate compound into ammonia (NH3). Then it is captured by hydrochloric acid in the Erlenmeyer flask. Distillation ends when the ammonia is perfectly distilled, indicated by the distillation results no longer being alkaline by checking using red litmus paper.

Data Analysis using Statistics

Nitrogen, N-Protein, total protein and actual protein in each sample were analyzed using t-test standard deviation method. Standard deviation was calculated using Formula 4.

$$SD = \sqrt{\frac{\sum(X-\bar{x})^2}{n-1}} \quad (4)$$

Data was rejected if t value ≥ table at the confidence interval of 99% (α = 0,01). t value was calculated using Formula 5.

$$t_{value} = \left|\frac{X-\overline{X}}{SD\sqrt{n}}\right| \quad (5)$$

Where SD is standard deviation, X is nitrogen, N-protein, total protein and actual protein, is mean of the nitrogen, N-protein, total protein and actual protein and n is number of determinations.

The nitrogen, N-protein, total protein and actual protein content was calculated using the Formula 6.

$$\mu = \overline{X} \pm t_{tabel} \times \frac{SD}{\sqrt{n}} \quad (6)$$

where, $\overline{X}$ is mean of the Nitrogen, N-Protein, total protein and actual protein, SD is standard deviation and n corresponds to number of determinations (Katoch, 2011; Sudarmadji et al., 1984).

Results and Discussion

The results of the analysis of total nitrogen (N), nitrogen-protein (N-Protein), total protein, actual protein, and non-protein nitrogen (NPN) content in fresh durian seeds are summarized in Table 1.

Table 1. Protein, Total Protein, Protein Actually and NPN in fresh durian seeds

Sample	N (g/100g)	N-Protein (g/100g)	Total Protein (g/100g)	Actual Protein (g/100g)	NPN (g/100g)
Bintana	0.62 ± 0.03a	0.52 ± 0.03a	3.88 ± 0.06a	3.25 ± 0.11a	0.98 ± 0.05a
Pelangi	0.45 ± 0.02b	0.36 ± 0.01c	2.81 ± 0.06b	2.25 ± 0.04c	0.81 ± 0.02b
Bawor	0.35 ± 0.01c	0.41 ± 0.01b	2.06 ± 0.06c	2.56 ± 0.07b	0.76 ± 0.02c

The total nitrogen (N), N-Protein, total protein, actual protein, and NPN content in durian seeds were affected by the moisture content of the seeds, which was approximately 30%. This moisture likely contributed to the lower levels of protein and nitrogen compounds observed across the different varieties of durian seeds.

For the Bintana variety, the total N content was 0.62 g/100 g, with N-Protein at 0.52 g/100 g, total protein at 3.88 g/100 g, NPN at 0.98 g/100 g, and actual protein at 3.25 g/100 g. The Pelangi variety contained 0.45 g/100 g of N, 0.36 g/100 g of N-Protein, 2.81 g/100 g of total protein, 2.25 g/100 g of actual protein, and 0.81 g/100 g of NPN. The Bawor variety exhibited the lowest values, with 0.35 g/100 g of N, 0.41 g/100 g of N-Protein, 2.06 g/100 g of total protein, 2.56 g/100 g of actual protein, and 0.76 g/100 g of NPN.

The observed decline in protein and nitrogen content is likely due to the exposure of the durian seed samples to environmental factors such as air, water, and sunlight. Extended contact with these elements can lead to protein denaturation, which involves the disruption of the peptide bonds and hydrophobic interactions within protein molecules, ultimately resulting in the breakdown of the protein structure. Sunlight and water likely contributed to these denaturation processes, further reducing the protein and nitrogen content in the seeds.

Notably, Bintana durian seeds exhibited higher values for total N, N-Protein, total protein, actual protein, and NPN compared to Pelangi and Bawor varieties. Similarly, Pelangi seeds had higher protein and nitrogen levels than Bawor seeds. Despite these variations, durian seeds, especially the Bintana variety, can still serve as a valuable source of nutrients, including carbohydrates, fats, and minerals.

In terms of nitrogen content, Bintana seeds had 0.17 g/100 g more total nitrogen compared to Bawor seeds, and Pelangi seeds contained 0.27 g/100 g more nitrogen than Bawor seeds. These findings suggest that different durian seed varieties may offer varying levels of nutritional value, which could be further explored for their potential uses in food or industrial applications.

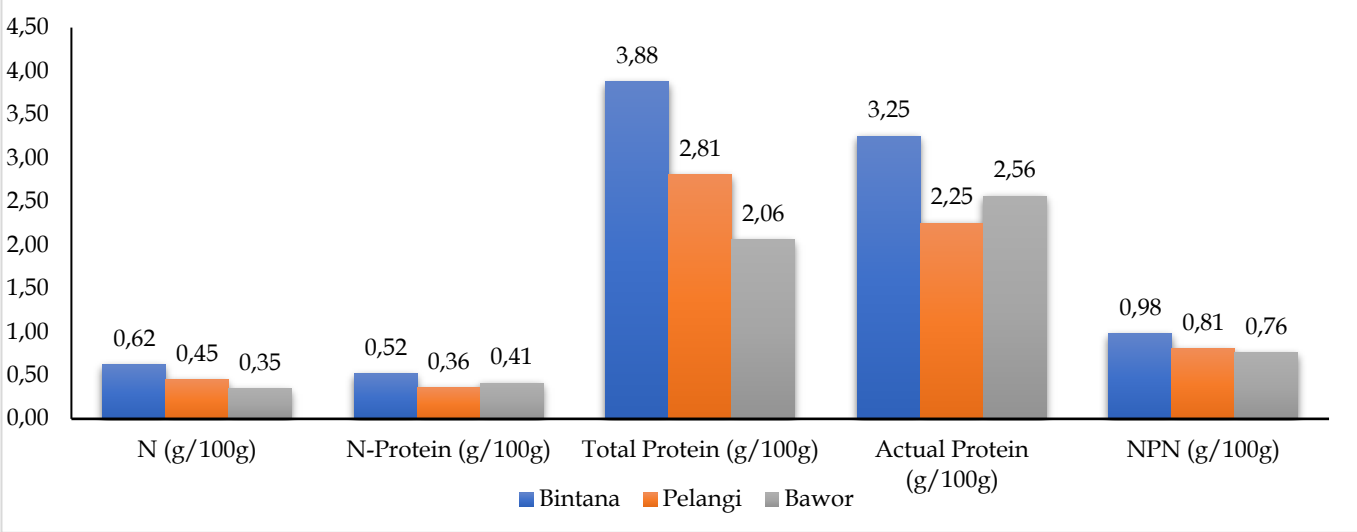

Figure 1. N-total, N-Protein, total protein, protein actually and NPN in fresh durian seeds

The results of the study on the examples used, Bintana, Pelangi, and Bawor durian seeds, can be seen in Table 2.

Table 2. Protein content of total N, N-Protein, total protein, actual protein decreased due to Bintana, Pelangi, and Bawor durian seeds.

Sampel	Rate (gr/100 g)				
	N-total	N-Protein	Total Protein	Actual Protein	NPN
Bintana	1.81±0.05a	0.87±0.02b	5.06±0.02a	4.17±0.06a	1.34±0.05b
Pelangi	1.75±0.04ab	1.54±0.04a	4.69±0.01b	3.38±0.06b	1.46±0.05a
Bawor	1.74±0.03b	1.53±0.04a	4.62±0.04c	3.31±0.08b	0.81±0.02c

Table 2. It can be seen that Bintana flour, that the N content is 1.81 gr/100 gr, N-Protein 0.87 gr/100 gr, total protein 2.06 gr/100gr, and the actual protein is 4.17 gr/100 gr and NPN 1.34 gr/100gr.

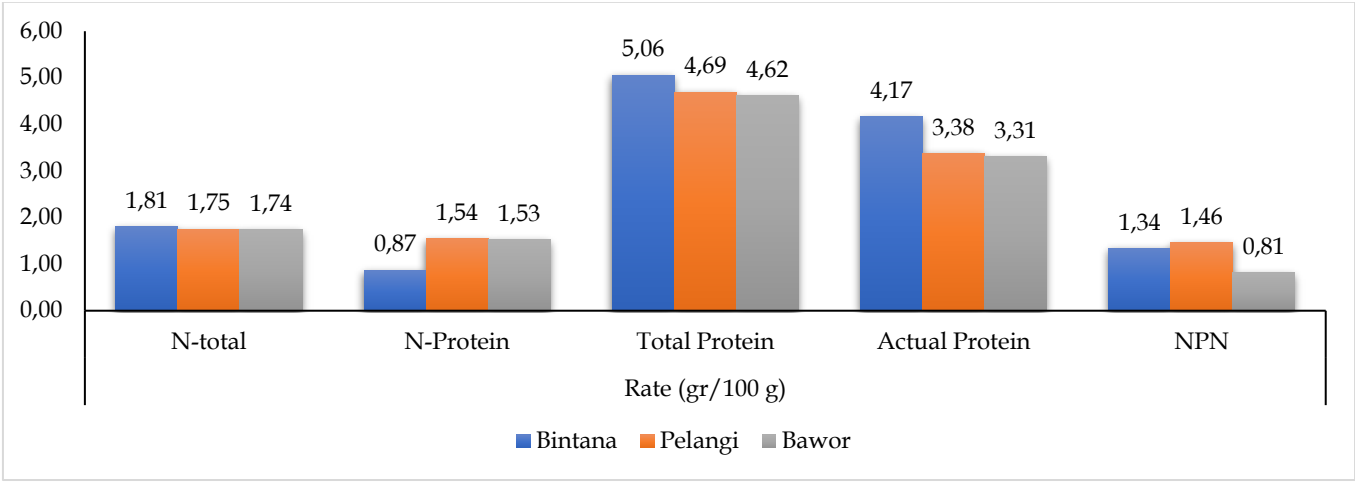

Figure 2. Protein content of total N, N-Protein, Total protein, actual protein decreased due to Bintana, Pelangi, and Bawor durian seeds.

Rainbow flour that the N content is 1.75 gr/100 gr, N-Protein 1.54gr/100 gr, total protein 4.69 gr/100gr gsn NPN 1.46 gr/100gr, Bawor flour that the N content is 1.74 gr/100 gr, N-Protein 1.53 gr/100 gr, total protein 4.62 gr/100gr, actual protein 3.31 and NPN 0.81. The total protein of bintana durian seeds is higher than that of pelangi and bawor durian seeds, there is a decrease in total protein and N-tolal due to the washing stage with

running water and soaking in hot water for 2 hours to remove mucus, then dried for 24 hours using an oven.

The protein content of fresh durian seeds is 10.1%, the decrease in protein content is very significant, but the decrease in protein content compared to fresh seeds is caused by heat because at the time of making flour there is soaking in water to remove mucus and also deactivate organic compounds, sunlight and protein dentauration occurs, peptide bonds, hydrophobic bonds and the opening of molecular bonds or polypeptide chains of protein molecules and affect total N, N-Protein, Total protein, actual protein and NPN. there is a difference between bintana flour and pelangi flour N-total of 0.06, and N total of 0.67 gr/100 gr, total protein of 0.37 gr/100 gr, actual protein of 0.78 gr / 100 and Bawor flour is higher than bintana NPN of 0.12 gr/100gr, Rainbow flour is higher than bawor flour the difference for N-total of 0.01 gr/100gr, N-Protein of 0.01 gr/100 gr, total protein of 0.07 gr/100 gr, actual protein of 0.07 gr/100 gr and NPN of 0.65 gr/100 g. The stages that occur during the destruction and distillation of the sample decomposition into its elements. Carbon, hydrogen elements are oxidized to CO, CO2 and H2O. Meanwhile, nitrogen will change into (NH4)2SO4. Distillation process. At this stage, the sample is heated in H2SO4 and broken down into $(NH_4)2SO_4$ by adding NaOH until it becomes a solution and is heated.

Conclusion

The analysis of protein content in fresh and processed durian seeds reveals significant differences across the three varieties: Bintana, Pelangi, and Bawor. Fresh Bintana seeds contained the highest levels of protein, with total nitrogen (N) at 0.62 g/100 g, N-Protein at 0.52 g/100 g, total protein at 3.88 g/100 g, actual protein at 3.25 g/100 g, and non-protein nitrogen (NPN) at 0.98 g/100 g. Pelangi seeds showed moderate protein content, with N at 0.45 g/100 g, N-Protein at 0.36 g/100 g, total protein at 2.81 g/100 g, actual protein at 2.25 g/100 g, and NPN at 0.81 g/100 g. Bawor seeds had the lowest values, with N at 0.35 g/100 g, N-Protein at 0.41 g/100 g, total protein at 2.06 g/100 g, actual protein at 2.56 g/100 g, and NPN at 0.76 g/100 g. This highlights the nutritional variability between durian seed varieties, with Bintana standing out as the most protein rich. Upon processing into flour, protein content increased across all varieties due to the processing steps, such as washing, soaking, and drying. Bintana flour showed the highest protein levels, with N at 1.81 g/100 g, N-Protein at 0.87 g/100 g, total protein at 5.06 g/100 g, actual protein at 4.17 g/100 g, and NPN at 1.34 g/100 g. Pelangi flour contained N at 1.75 g/100 g, N-Protein at 1.54 g/100 g, total protein at 4.69 g/100 g, actual protein at 3.38 g/100 g, and NPN at 1.46 g/100 g. Bawor flour showed slightly lower values, with N at 1.74 g/100 g, N-Protein at 1.53 g/100 g, total protein at 4.62 g/100 g, actual protein at 3.31 g/100 g, and NPN at 0.81 g/100 g. This demonstrates the impact of processing on enhancing the protein content in durian seeds, making them a more valuable nutritional source. In summary, durian seeds, especially those from the Bintana variety, exhibit substantial potential as a nutrient-rich ingredient. While fresh seeds display varying protein contents, the processed flour form shows significant protein enhancement, indicating that durian seeds, particularly Bintana, could be a promising source for various applications in food production and other industries.

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Conflicts of Interest

The authors declare no conflict of interest.

References

Ali, N. A. W. A., Wong, G. R., Tan, B. C., Lum, W. S., & Mazumdar, P. (2025). Unleashing the Potential of Durian: Challenges, Opportunities, and the Way Forward. *Applied Fruit Science*, *67*(1), 1–15. https://doi.org/10.1007/s10341-024-01237-y

Amid, B. T., Mirhosseini, H., & Kostadinović, S. (2012). Chemical composition and molecular structure of polysaccharide-protein biopolymer from Durio zibethinusseed: extraction and purification process. *Chemistry Central Journal*, *6*(1). https://doi.org/10.1186/1752-153x-6-117

Baqi, A., Lim, V., Yazid, H., Khan, F. . A., Lian, C. J., Nelson, B. R., Seelan, J. S. S., Appalasamy, S., Mokhtar, S. I., & Kumaran, J. V. (2021). A review of durian plant-bat pollinator interactions. *Journal of Plant Interactions*, *17*(1), 105–126. https://doi.org/10.1080/17429145.2021.2015466

Gunamantha, I. M., Oviantari, M. V, Suryaputra, I. G. N., Sudiana, I. K., Sastrawidana, I. D. K., Armatini, K., & Handayani, D. F. (2021). Nitrogen content and C-Organic in the field for tropical fruit plantation in Jinengdalem Village, Bali, Indonesia. *IOP Conference Series: Earth and Environmental Science*,

824(1), 12006. https://doi.org/10.1088/1755-1315/824/1/012006

Hamim, N. A., Saari, N., Wan Ibadullah, W. Z., Mohamed, A. M. Dos, Anwar, F., Hasan, M. Y., Abdul Rahim, M. Z., Abdul Rahman, M. S., & Karim, R. (2025). Optimization of extraction for efficient recovery of kenaf seed protein isolates: evaluation of physicochemical and techno-functional characteristics. *Journal of the Science of Food and Agriculture*, *105*(3), 2032–2047. https://doi.org/10.1002/jsfa.13979

Harvey, D. (1999). *Modern Analytical chemistry*. McGraw Hill. Retrieved from https://shorturl.asia/jg4ON

Husin, N. A., Rahman, S., Karunakaran, R., & Bhore, S. J. (2018). A review on the nutritional, medicinal, molecular and genome attributes of Durian (Durio zibethinus L.), the King of fruits in Malaysia. *Bioinformation*, *14*(6), 265. https://doi.org/10.6026/97320630014265

Ismail, B. P., & Smith, D. M. (2024). Protein Separation and Characterization Procedures. In *Nielsen's Food Analysis* (pp. 395–418). Springer. https://doi.org/10.1007/978-3-031-50643-7_24

Juan, C. A., de la Lastra, J. M., Plou, F. J., & Pérez-Lebeña, E. (2021). The chemistry of reactive oxygen species (ROS) revisited: outlining their role in biological macromolecules (DNA, lipids and proteins) and induced pathologies. *International Journal of Molecular Sciences*, *22*(9), 4642. https://doi.org/10.3390/ijms22094642

Katoch, R. (2011). Analytical techniques in biochemistry and molecular biology. In *Springer eBooks*. Springer eBooks. https://doi.org/10.1007/978-1-4419-9785-2

Khaksar, G., Kasemcholathan, S., & Sirikantaramas, S. (2024). Durian (Durio zibethinus L.): Nutritional Composition, Pharmacological Implications, Value-Added Products, and Omics-Based Investigations. *Horticulturae*, *10*(4), 342. https://doi.org/10.3390/horticulturae10040342

Kostryco, M., & Chwil, M. (2022). Nectar Abundance and Nectar Composition in Selected Rubus idaeus L. *Varieties. Agriculture*, *12*(8), 1132. https://doi.org/10.3390/agriculture12081132

Le, N. T. A. (2024). *Upcycling of fruit processing byproducts for enhancing food preservation and delivery*. Nanyang Technological University. Retrieved from https://dr.ntu.edu.sg/handle/10356/183067

Lin, D., Sun, L., Chen, Y., Liu, G., Miao, S., & Cao, M. (2022). Peptide/protein hydrolysate and their derivatives: Their role as emulsifying agents for enhancement of physical and oxidative stability of emulsions. *Trends in Food Science & Technology*, *129*, 11–24. https://doi.org/10.1016/j.tifs.2022.08.012

Morris, R., Black, K. A., & Stollar, E. J. (2022). Uncovering protein function: from classification to complexes. *Essays in Biochemistry*, *66*(3), 255–285. https://doi.org/10.1042/EBC20200108

Purnama, N., Said, I., Rahmawati, N. S., & Dandi, N. (2022). The use of durian seeds (Durio zibethinus Murr) as flour products from Tolitoli and Donggala Regencies. *Jurnal Pengelolaan Sumberdaya Alam Dan Lingkungan (Journal of Natural Resources and Environmental Management*, *12*(3), 478–484. https://doi.org/10.29244/jpsl.12.3.478-484

Retnoningsih, A., Rahayu, E. S., & Sari, I. P. (2016). Chacarterization of Local Durian Germplasm Based on The Morphology of Fruit. *Sainteknol: Jurnal Sains Dan Teknologi*, *14*(2), 89–94. https://doi.org/10.15294/sainteknol.v14i2.8989

Soto-Sierra, L., Stoykova, P., & Nikolov, Z. L. (2018). Extraction and fractionation of microalgae-based protein products. *Algal Research*, *36*, 175–192. https://doi.org/10.1016/j.algal.2018.10.023

Sudarmadji, S., Haryono, B., & Suhardi. (1984). *Prosedur Analisa untuk Bahan Makanan dan Pertanian*. Liberty. https://cir.nii.ac.jp/crid/1130000797278900992

Tongdee, S. (1992). Postharvest Handling and Technology of Tropical Fruit. *Acta Horticulturae*, *321*, 713–717. https://doi.org/10.17660/actahortic.1992.321.89

Van Hau, T., Tien, D. H., Thuy, N. M., Ky, H., Yen, T. T. O., Van Tri, M., Van Hoa, N., & Hieu, T. S. (2023). Durian. In *Tropical and Subtropical Fruit Crops* (pp. 161–200). Apple Academic Press. https://doi.org/10.1201/9781003305033-3

Wang, D., Eraslan, B., Wieland, T., Hallström, B., Hopf, T., Zolg, D. P., Zecha, J., Asplund, A., Li, L., Meng, C., Frejno, M., Schmidt, T., Schnatbaum, K., Wilhelm, M., Ponten, F., Uhlen, M., Gagneur, J., Hahne, H., & Kuster, B. (2019). A deep proteome and transcriptome abundance atlas of 29 healthy human tissues. *Molecular Systems Biology*, *15*(2). https://doi.org/10.15252/msb.20188503

Wiangsamut, B., & Wiangsamut, M. E. L. (2023). Assessment of natural fruit ripening and fruit quality of three elite durian cultivars for overland export. *Trends in Sciences*, *20*(5), 4647. https://doi.org/10.48048/tis.2023.4647

Yao, Y., Li, J., Ma, K., Zhang, J., Lin, Y., Tan, H., Yu, J., & Fang, F. (2024). Characteristics of Dissolved Organic Matter (DOM) Combined with As in Fe-Rich Red Soils of Tea Plantations in the Southern Anhui Province, East China. *Agriculture*, *14*(12), 2289. https://doi.org/10.3390/agriculture14122289

Zhou, H., & Pang, X. (2018). Electrostatic interactions in protein structure, folding, binding, and condensation. *Chemical Reviews*, *118*(4), 1691–1741. https://doi.org/10.1021/acs.chemrev.7b00305