

Exploring the Viability of Waste Durian (*Durio zibethinus*) Seeds and Cempedak (*Artocarpus integer*) Seeds as Nutritive Resources: An Analytical Study of Their Suitability via Alcohol and Glucose Assays

Syarpin¹, Rizki Rachmad Saputra^{2*}, Nur Haliza¹, Anista¹, Helda Yanti¹, Tahsya Amanda Sisilya¹, Rifaldi Lutfi Fahmi³

¹Department of Chemistry Education, Faculty of Education and Teacher Training, Universitas Palangka Raya, Palangka Raya 74874, Central Kalimantan, Indonesia

²Department of chemistry, Faculty of Mathematics and Science, Universitas Palangka Raya, Palangka Raya 74874, Central Kalimantan, Indonesia

³Department of Food Science and Nutrition, Poznań University of Life Sciences, Poland

Abstract

Durian (*Durio zibethinus*) and Cempedak (*Artocarpus integer*), prominent staples in Indonesia, harbor seeds that can be harnessed for the production of 'Tapai', a traditional fermented product. This fermentation process within cempedak and durian seeds manifests in the generation of alcohol and glucose at discernible thresholds. Motivated by the prevalent underutilization of durian and cempedak seeds leading to wastage, this study aims to assess the viabilities of Tapai derived from these seeds by scrutinizing their alcohol and glucose profiles. Employing a quantitative descriptive approach, the research centers on cempedak and durian seed Tapais as the primary samples. Focusing on variables like alcohol and glucose content, the investigative technique incorporates distillation and titration using the Luff Schoorl method. Results are subsequently organized in frequency distribution tables following coding and tabulation. Observations reveal that alcohol and glucose levels in cempedak seed Tapai on the 10th day stand at 0.00% and 30.05%, respectively, while on the 15th day, they escalate to 4.88% and 41.92%. In the case of durian seed Tapai, levels on the 10th day are recorded at 4.88% for alcohol and 36.90% for glucose. On the 15th day, these figures rise to 16.57% for alcohol and 46.53% for glucose. Ultimately, this study underscores that the alcohol and glucose concentrations in cempedak and durian seed Tapais experience progressive augmentation over a 5-day period, rendering them safely consumable in moderate quantities.

Keywords: *fermentation, alcohol content, glucose level, cempedak seed tapai, durian seed tapai*

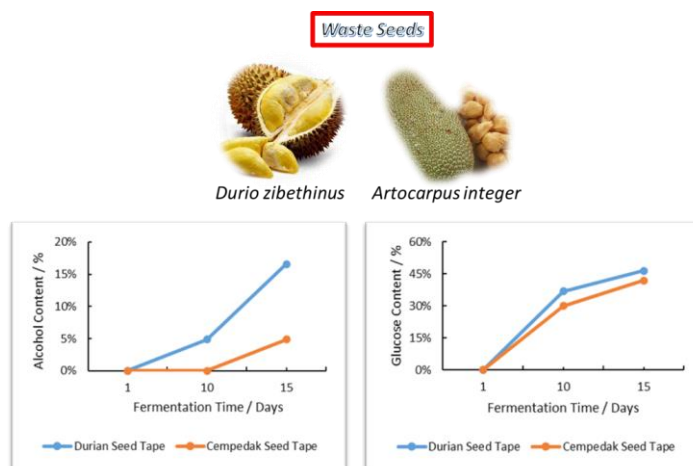
* Corresponding author
Email addresses: rizkirachmads@mipa.upr.ac.id

DOI: [10.22437/chp.v7i2.27983](https://doi.org/10.22437/chp.v7i2.27983)

Received 28 August 2023; Accepted 30 December 2023; Available online 31 December 2023

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Graphical Abstract



Introduction

Indonesia is home to a myriad of beloved fruits like durian, oranges, bananas, pineapples, guavas, jackfruits, cempedak, and others ^[1]. These fruits are currently becoming widely distributed and marketed in addition to being produced in large quantities. Remarkably, the by-products also contain great nutritional and functional potential, especially the seeds and peels, which make up 10–35% of the fruit weight. Yet, the potential lies in reutilizing these byproducts, transforming them into valuable commodities for the community ^[1,2]. For instance, cempedak peels can undergo processing, resulting in a delectable side dish, while durian and cempedak seeds can be easily boiled to create uncomplicated food preparations ^[3].

As evidenced by 2020 data from the Central Bureau of Statistics, Palangka Raya witnessed fruit production figures of 6,234 tons for durian and 13,652 tons for cempedak in a single year ^[4]. These numbers underscore the escalating volume of discarded durian and cempedak seeds if left untreated. It's worth noting that both durian and cempedak seeds boast inherent benefits. Cempedak seeds, rich in carbohydrates and energy, offer a valuable nutritional source. Similarly, durian seeds, boasting a substantial carbohydrate content of 43.2 g per 100 grams, hold significant potential as viable food ingredients ^[5].

The process of fermentation involves utilizing microbial metabolism to transform raw materials into more valuable end products, such as organic acids, single-cell proteins, antibiotics, and biopolymers ^[6]. This approach gives rise to a range of fermented goods, including oncom, tempeh, sufu, soy sauce, angkak, cheese, wine, kefir, yogurt, and Tapai. In Indonesia, Tapai stands as a popular snack, commonly available in two variants: Tapai ketan and Tapai cassava ^[7]. To enhance Tapai's consumer base and simultaneously minimize waste from durian and cempedak seeds, an innovative avenue emerges in the realm of Tapai production. A potential strategy involves substituting the traditional core components of Tapai with durian seeds and cempedak seeds, thereby offering a novel variation that aligns with evolving preferences and resource optimization ^[8].

The making of Tapai is inseparable from the role of yeast in its fermentation. Microorganisms in yeast will convert complex compounds into simple compounds such as zymase enzymes produced by microorganisms of the genus *Saccharomyces* convert glucose into alcohol ^[9]. Some types of microorganisms contained in yeast are *Chlamy domucor oryzae*, *Rhizopus oryzae*, *Mucor sp.*, *Candida utilis*, *Saccharomyces cerevicae*, *Saccharomyces verdomanii*. The protein and flavor of Tapai produced is determined by the type of microorganisms that are active in yeast ^[10]. The aforementioned yeast, including *Saccharomyces cerevisiae*, commonly employed

in fermentation, has the capability to decompose carbohydrates into basic glucose units. Moreover, with the aid of the invertase enzyme found in the yeast, these uncomplicated sugar units can be transformed into ethanol [11]. Based on the description above, the researcher conducted a study on the Utilization of Durian Seed and Cempedak Seed Waste as Food Ingredients through Alcohol Content Test and Glucose Content Test.

Experimental Section

Materials

Durian seeds, Cempedak seeds, Yeast, Aquadest, Banana leaves, Ice cubes, KI (Merck), H₂SO₄ (Sigma Aldrich), Sodium thiosulfate (Sigma Aldrich), Luff Schoorl solution.

Instrumentation

Aluminum foil, Amylum, Gas stove (maspion), Pan, Beaker Glass (Duran), Distillation flask (pyrex), Pycnometer (Iwaki), Analytical balance, Mortar, Bowl, Filter paper, Erlenmeyer (pyrex), Spatula, burette (Duran), Measuring flask (iwaki), Volumetric pipette (duran). Hot plate (IKA), Jar.

Procedure

Material preparation

Preparation of durian seeds and cempedak seeds commenced with 500 grams of each variety, subjected to meticulous cleaning through thorough washing. Subsequently, the immaculate seeds underwent a cooking process until reaching optimal tenderness. Once cooked, the seeds were carefully drained and allowed to cool. Following this, the outer coverings of both durian and cempedak seeds were meticulously removed until achieving a pristine state. The seeds then underwent an additional cleansing phase, ensuring their utmost cleanliness. To instigate the fermentation process, a measured quantity of approximately 1 gram of crushed Tapai yeast was evenly dispersed onto the surfaces of each individual durian and cempedak seed. These treated seeds were thoughtfully enwrapped within banana leaves, imparting a traditional touch to the process. Finally, the seeds, encased in their leafy wraps, were securely stored within sealed containers,

providing an environment conducive to the ensuing fermentation journey.

Alcohol content assay

Commence by precisely measuring 200 grams of both durian and cempedak seed Tapai. Subsequently, proceed to meticulously grind the seeds until a uniformly smooth texture is achieved. Introduce 200 ml of distilled water into this mixture. The concocted blend is then carefully transferred into a beaker, where it is vigorously agitated to ensure thorough homogeneity. Utilize filter paper to effectively partition the mixture, with the resultant filtrate meticulously poured into a distillate flask. This distillate flask should be firmly linked to the distillation setup, which is meticulously configured to uphold temperatures within the range of 78-100°C. Upon completion of the distillation process, the distilled product is meticulously gathered within a 200 ml erlenmeyer flask. Subsequently, meticulously place the distilled sample into a pycnometer that has been meticulously weighed beforehand. Utilizing an analytical balance, ascertain the specific gravity (SG) of the sample with precision, aligning these observations with the relevant alcohol content reference table [12].

Glucose content assay

The Luff Schoorl method begins with measuring 2.5 grams of both durian and cempedak seed Tapai in order to calculate the glucose concentrations. The samples are then diluted to reduce concentration in 100 mL volumetric flasks. Dilution is followed by extraction of the filtrate. After dilution, the sample solution is heated for 30 minutes to speed up the reaction. Following this 2-minute heating cycle, the solution is mixed with Luff reagent before undergoing one more heating cycle. Ice is used to cool the mixture, and then 20% KI solution and 6N H₂SO₄ solution are added. Titration continues using 0.1N Na₂S₂O₃ until the color turns a light shade of brown. After that, 1% amyllum solution is added, and titration is carried out until the color turns milky white. Subsequently, the glucose content within durian seed Tapai and cempedak seed Tapai is analyzed through the subsequent formula [13]:

$$\% \text{ Glucose} = \frac{W_{\text{glucose}} \times Df}{W_{\text{sample}}} \times 100\% \dots\dots\dots [1]$$

Where W glucose is weight of reduced glucose, Df is dilution factor, and W sample is weight of sample.

Results and Discussions

Sample preparation

The process of crafting Tapai from durian seeds and cempedak seeds mirrors the conventional Tapai-making process. Both durian seed Tapai and cempedak seed Tapai are fashioned from the fundamental components of durian seeds and cempedak seeds, which are subjected to boiling until they attain full tenderness. This procedure, termed hydrolysis reaction, effectively breaks down starch into glucose [14]. Subsequently, the outer layers of durian seeds and cempedak seeds are peeled away. This sets the stage for the fermentation process, wherein a uniform distribution of the mixture is applied onto the seeds of both durian and cempedak. Following this, the seeds are meticulously enclosed within an airtight container, enveloped in banana leaves, to ensure an anaerobic environment [15]. This controlled fermentation process leverages yeast, specifically *Saccharomyces cerevisiae*, within a predefined timeframe.

Maintaining a sealed environment is critical during fermentation, as the presence of oxygen could undermine the process, leading to incomplete fermentation of cempedak and durian seeds. Yeast orchestrates the conversion of carbohydrates within these seeds into alcohol and water. The incorporation of alcohol imparts a sweet and slightly tart flavor to cempedak and durian seeds. The yeast quantity employed corresponds to the volume of cempedak and durian seeds used. The outcome of fermentation culminates in durian seed Tapai and cempedak seed Tapai, distinguished by their unique aroma, tender consistency, and subtly sweet and sour taste [16]. To gauge the effects of fermentation, the resulting products are subjected to alcohol and glucose content assessment on both the 10th and 15th days, elucidating the magnitude of changes over a 5-day interval.

Distillation-based technique of determining alcohol content

The The specific gravity of the sample was measured utilizing a pycnometer to ascertain its alcohol content. In accordance with Regulation No: 86/MenKes/Per/IV/77, alcoholic beverages, including liquors, are distinct from drugs. The categorization is as follows: beverages containing 1-5% alcohol are classified as class A liquors, those with alcohol content $\geq 20\%$ fall under class B liquors, and beverages with alcohol content $\geq 55\%$ are categorized as class C [17].

The determination of alcohol content in fermented cempedak and durian seed Tapai using the pycnometer method provides valuable insights into their classification and safety for consumption, aligning with international regulations and guidelines. The study's adherence to Regulation No: 86/MenKes/Per/IV/77 underscores the importance of defining clear distinctions between alcoholic beverages and drugs, categorizing alcoholic beverages based on their alcohol content into class A, B, and C liquors. The analysis demonstrates that the cempedak seed Tapai falls within the class A liquor category, containing alcohol content ranging from 1-5%. This finding substantiates its safety for consumption, aligning with international standards. Similarly, the freshly made durian seed Tapai also falls within the class A category, affirming its safe consumable status. However, the study reveals an interesting transformation for durian seed Tapai over time. As fermentation progresses, its alcohol content increases, transitioning it into the class B liquor category ($\geq 20\%$ alcohol content) after aging [18]. While still considered safe for consumption within recommended limits, this shift highlights the evolving nature of alcoholic content during the fermentation process and its implications for consumer awareness [19].

Based on the derived alcohol content, the cempedak seed Tapai aligns with class and is deemed safe for consumption. Meanwhile, the freshly made durian seed Tapai also falls into class A, representing a safe consumable, but upon aging, it transitions to class B, still within safe consumption limits yet not recommended in

excessive amounts. The analysis of alcohol content in cempedak seed Tapai upon completion on the 10th day revealed a negligible 0.00%. After allowing it to stand for 5 days, the alcohol content increased to 4.88%. Conversely, the alcohol content in durian seed Tapai upon completion on the 10th day registered at 4.88%, escalating to 16.57% after 5 days more of fermentation (Table 1, Figure 1).

The observed changes in alcohol content during fermentation unveil intriguing dynamics within the Tapai products. The increase in alcohol content over time is a result of the ongoing microbial activity, which metabolizes sugars into alcohol. This phenomenon contributes to the flavor and overall profile of fermented foods, enriching our understanding of the complex biochemical transformations occurring during the fermentation process [20]. This research contributes to the broader discourse on fermented foods and their safety considerations. The findings underscore the importance of monitoring alcohol content as an integral aspect of product quality control and consumer safety. Furthermore, the study's approach to classification based on alcohol content aligns with global efforts to standardize definitions and regulations surrounding alcoholic beverages. The research also highlights the significance of traditional fermented foods in cultural and culinary contexts. By providing evidence of the evolution of alcohol content in durian seed Tapai, the study underscores the need for informed consumption practices and reinforces the importance of moderating consumption of fermented products, particularly those that

transition to higher alcohol content categories with extended aging.

The meticulous analysis of alcohol content in cempedak and durian seed Tapai showcases the relevance of precise measurement techniques and adherence to regulatory frameworks in ensuring consumer safety. The study's findings, which align with international classification standards, not only contribute to scientific knowledge but also resonate with broader discussions on food safety, fermentation, and cultural gastronomy. This research serves as a stepping stone for further investigations into the intricate interplay between fermentation, alcohol content, and the sensory qualities of traditional fermented foods within a global context.

According to Fajria et al.'s earlier research, A. champeden seed carbohydrates can reach up to 38.16% [21]. Purnama et al., 2022, on the other hand, showed that durian seed contains 56.16% carbs [22]. The studies conducted by Fajria et al. and Purnama et al. revealed crucial connections between carbohydrate content and its impact on alcohol production in cempedak and durian seeds. Cempedak seeds, containing up to 38.16% carbs, and durian seeds, with a higher content of 56.16%, likely explain observed variations in alcohol production during fermentation. Fermentation dynamics highlight this, with cempedak seed Tapai exhibiting minimal alcohol (0.00%) on the 10th day, indicating a slower process. In contrast, durian seed Tapai has 4.88% alcohol on the 10th day, rising to 16.57% after 5 more days.

Table 1. Alcohol and glucose content of durian and cempedak seed tapai.

Tapai	Fermentation time (Days)	Alcohol Content	Glucose Content
Durian Seed	1	0.00	0.00
	10	4.88	36.10
	15	16.57	46.53
Cempedak Seed	1	0.00	0.00
	10	0.00	30.05
	15	4.88	41.92

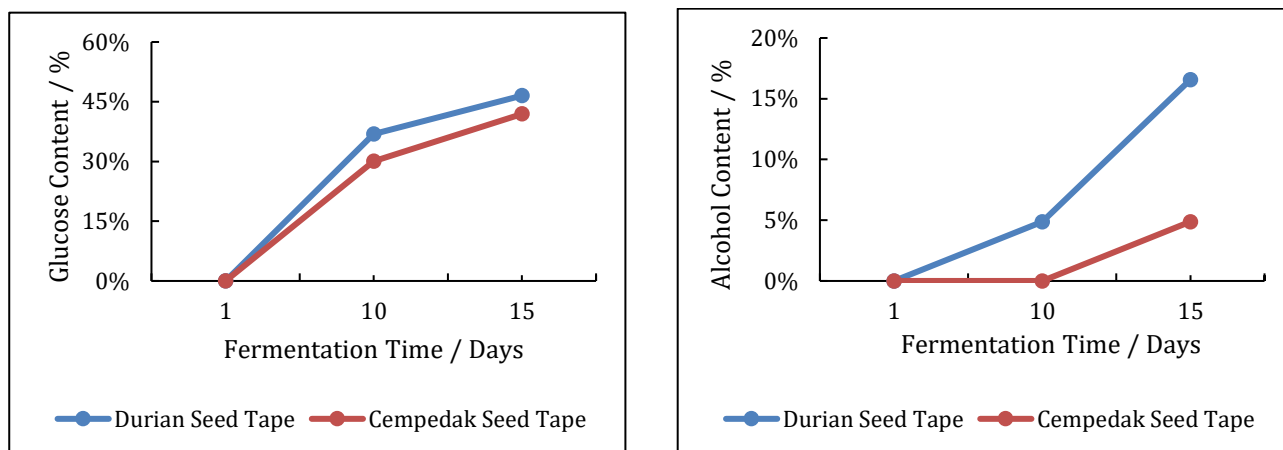


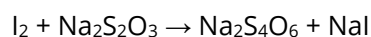
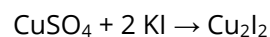
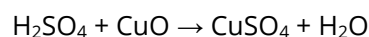
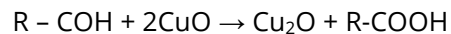
Figure 1. Increase in alcohol and glucose content of durian seed and cempedak seed Tapais.

This fluctuation in alcohol content may be attributed to disparities in the carbohydrate and nutrient composition of cempedak and durian seeds. Recognizing the crucial role of nutrition in fermentation is essential, especially with the higher carbohydrate content in durian seeds, potentially leading to a more significant increase in alcohol. Understanding seed nutritional composition is critical for predicting and controlling the fermentation process. This knowledge extends beyond the lab, benefiting the food processing industry, providing insights for creating fermented products with specific characteristics [20].

The varying carbohydrate content between cempedak and durian seeds not only emphasizes diverse nutritive resources in different fruits but also suggests implications for consumer preferences in fermented products, including flavor and nutritional content. In essence, the correlations from the research offer insights into the carbohydrate composition of cempedak and durian seeds, providing opportunities to understand nutritive resources and explore applications of fermented products [17]. These findings significantly contribute to emphasize the importance of considering seed nutritional composition in optimizing the fermentation process for desired outcomes.

Measure the glucose content of durian seed Tapai and cempedak seed Tapai by The Luff School method

The reaction between monosaccharides and copper solution is the basis for this technique. The following can be stated as the reaction that takes place during Luff School's determination of sugar [22]:



The durian seed Tapai's glucose content was 36.90% on the 10th day of fermentation and increased to 46.53% on the 15th. On the 10th day of fermentation, the cempedak seed Tapai's glucose content was 30.05%; on the 15th day, it had increased to 41.92% (Table 1, Figure 1). The length of fermentation is directly related to the rise in glucose levels in both durian seed Tapai and cempedak seed Tapai. Higher glucose levels in the Tapai are correlated with longer fermentation times [23].

The Luff School method presents a comprehensive and meticulous approach for quantifying glucose concentrations in durian and cempedak seed Tapai samples.

The method encompasses several essential steps, including initial measurements, dilution, extraction, and a series of heating and chemical reactions. By following this systematic procedure, that step is able to accurately assess the glucose content within the samples. The two-step heating process, combined with the incorporation of Luff reagent, KI solution, and H₂SO₄ solution, optimizes the chemical reactions that facilitate the determination of glucose levels. The titration process using Na₂S₂O₃ is a critical stage, during which the color transitions from a light brown shade to a milky white appearance, indicating the completion of the reaction. This comprehensive methodology not only ensures accuracy but also provides a clear visual endpoint for researchers to identify ^[24].

The obtained results from the study exhibit a distinct pattern of glucose concentration changes during the fermentation process of durian and cempedak seed Tapai. The observed increase in glucose content over time reveals a direct correlation between the length of fermentation and the rise in glucose levels ^[25]. This correlation underscores the significance of fermentation duration in shaping the glucose composition of Tapai products. Moreover, the study's findings have broader implications for the field of food science and fermentation. The Luff School method can serve as a valuable tool for researchers and practitioners seeking to assess the biochemical changes occurring in fermented foods. The precise quantification of glucose levels not only contributes to our understanding of fermentation dynamics but also opens avenues for process optimization and quality control within the food industry ^[24].

This research enriches the knowledge base related to traditional fermented products, particularly from Southeast Asia. By presenting a well-defined method for analyzing glucose content, the study bridges the gap between traditional practices and modern analytical techniques. Furthermore, the study's emphasis on the relationship between fermentation time and glucose levels can inspire further investigations into the underlying biochemical mechanisms driving these changes. The Luff School method, outlined in this study, offers a

robust framework for the assessment of glucose concentrations in durian and cempedak seed Tapai ^[26]. The method's thorough approach, supported by clear experimental results, has the potential to reshape our understanding of fermentation processes and their impact on the composition of traditional foods. This research stands as a valuable contribution to the global discourse on food science and fermentation, fostering deeper insights and encouraging future explorations in the realm of culinary traditions and scientific innovation.

Conclusions

In conclusion, the conducted research highlights a significant correlation between yeast-driven fermentation duration and the subsequent elevation in alcohol levels and glucose content within the Tapai production process. The results unequivocally demonstrate that extended fermentation periods contribute to intensified alcohol production and glucose accumulation. This phenomenon is particularly evident in the case of durian seeds, which exhibited the highest alcohol content (16.57%) and the most elevated glucose level (46.53%). Conversely, cempedak seeds showcased comparatively lower alcohol contents (4.88%) alongside glucose levels of 41.92%. These findings emphasize the potential of harnessing durian and cempedak seeds as valuable nutritive resources through the fermentation process. Not only do these seeds offer the opportunity to create delectable Tapai products, but the varying alcohol and glucose profiles also present avenues for diversifying and optimizing the utilization of these underutilized waste materials. This insight into their transformation underscores the feasibility of repurposing these seeds as valuable sources of both taste and nutrition, aligning with sustainable practices and innovative approaches to reduce waste and enhance nutritional outcomes.

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