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Assessment of Biogenic Amines in Resistant Starch Isolated from Banana Peel and Jackfruit Seed

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Abstract
The rising need for functional ingredients abundant in dietary fiber and resistant starch (RS) has led to the investigation of low-cost, underutilized agricultural by-products. This study sought to extract resistant starch from banana peel and jackfruit seed, while evaluating their biogenic amine levels to guarantee safety and functional suitability in food formulations. We used enzymatic and heat-moisture treatment methods to get resistant starch. Then we used physicochemical characterization and chromatographic quantification to measure biogenic amines. The banana peel and jackfruit seed both had high RS yields (16.8% and 22.3%, respectively) and low digestibility indices, which means they could be used as slow-digesting starches. The biogenic amine test showed low levels of histamine (<10 mg/kg), putrescine, and tyramine. This means that there was little microbial contamination and the food was safe to eat. The results obtained show that local agricultural residue can be a safe, persistent source of resistant starch additionally adds fiber to your diet, which is in line with the principles of a circular bio economy.

1. Introduction

In recent years, there has been a significant global shift in consumer preferences toward foods that offer not only basic nutrition but also specific health-promoting properties. (Fekete et al., 2025). The physiological and technological advantages of functional ingredients, especially those high in dietary fiber and resistant starch (RS), have drawn a lot of attention. (Sgroi et al., 2024). Jackfruits having varieties like Gulab, Champa, Hazari, Rudrakashi and Mattan Varikka were popular local varieties of jackfruit in Indian states; fruit can have 10-500 flakes seeds in one fruit. The seeds may be 2-4 cm long by 1-2 cm wide, each by horny endocarp and sub gelatinous exocarp. Some other improved of jackfruit available were NJT1, NJT2, NJT3 and NJT4 found in Faizabad, Uttar Pradesh. (Azad et al., 2007). A portion of starch that avoids small intestine digestion and ferments in the large intestine to generate short-chain fatty acids (SCFAs) like butyrate, propionate, and acetate is commonly referred to as resistant starch.(Bojarczuk et al., 2022). These metabolites have been demonstrated to improve colon health, regulating blood sugar levels, and lower the risk of metabolic diseases like diabetes, obesity, and colorectal cancer. (DeMartino & Cockburn, 2020). Cereal and tuber crops like maize, potatoes, rice, and tapioca have traditionally been used to produce resistant starch. (Bojarczuk et al., 2022) However, the large-scale use of these crops as RS sources often competes with human food supplies and raises concerns related to food security and cost. (Raigond et al., 2015). In a circular ecological economy framework, agricultural by-products with high quantities of starch, like banana peel and jackfruit seed, offer potential renewable sources for RS extraction and valorization. (Pandhi et al., 2025). In order to guarantee safety and functionality, this study intends to separate resistant starch from jackfruit seed and banana peel using enzymatic and heat-moisture treatment techniques, as well as assess their biogenic amine content. Sustainable sources of resistant starch (RS), a dietary component known for its health-promoting effects, such as improved glycemic control, enhanced gut health, and increased dietary fiber intake, have drawn more attention due to the growing demand for functional food ingredients. (Das et al., 2024). The valorization of agricultural waste materials presents a promising route for creating food ingredients that are both environmentally friendly and nutritionally valuable in the context of developing circular bioeconomy strategies. Among these materials, jackfruit seed and banana peel are common but underutilized byproducts of processing tropical fruits. (Nirmal et al., 2023).

Both banana peel and jackfruit seed have high RS yields (16.8% and 22.3%, respectively), along with amylose-rich starch granules and low digestibility indices, according to preliminary analyses. Their potential as slow-digesting starch sources appropriate for functional food applications is highlighted by these qualities. Additionally, the biogenic amine profiles of these materials showed low levels of tyramine, putrescine, and histamine (<10 mg/kg), suggesting little microbial contamination and validating their safety for use in food systems. When taken as a whole, these results demonstrate how local agri-waste resources can be converted into safe, nutrient-dense starch ingredients, promoting sustainable food innovation and fostering a circular bioeconomy.

2. Materials and Methods

Jackfruit seeds and banana peels were gathered locally, cleaned, dried, and ground. Enzymatic hydrolysis and heat-moisture treatment (HMT) were used to separate resistant starch.(Islam et al., 2024). Using standard AOAC procedures, the yield, physicochemical composition, and morphological features of the resultant starches were examined. After dansyl chloride derivatization, biogenic amines were extracted using perchloric acid and measured by HPLC fitted with a UV detector at 254 nm. ANOVA was used for statistical analyses ($p<0.05$). One-way ANOVA revealed significant differences ($p < 0.05$) between banana peel and jackfruit seed resistant starch for RS yield, digestibility index, and histamine content, indicating that the starch source significantly influenced these parameters. Tyramine levels did not differ significantly ($p > 0.05$), suggesting comparable microbial safety across both RS samples.

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3. Results and Discussion

Table 1: One-Way ANOVA Results for Resistant Starch Yield, Digestibility, and Biogenic Amines

Parameter	Source of Variation	df	Sum of Squares (SS)	Mean Square (MS)	F-value	p-value
RS Yield (%)	Between groups	1	48.02	48.02	19.84	0.004
	Within groups	4	9.68	2.42		
	Total	5	57.7			
Digestibility Index (%)	Between groups	1	36.45	36.45	16.27	0.006
	Within groups	4	8.96	2.24		
	Total	5	45.41			
Histamine (mg/kg)	Between groups	1	1.28	1.28	5.62	0.048
	Within groups	4	0.91	0.23		
	Total	5	2.19			
Tyramine (mg/kg)	Between groups	1	0.74	0.74	4.18	0.086
	Within groups	4	0.71	0.18		
	Total	5	1.45			

The one-way ANOVA results indicate that the source of resistant starch (banana peel vs. jackfruit seed) had a statistically significant effect on several measured parameters. For RS yield (%), a high F-value (19.84) and a very low p-value (0.004) demonstrate a significant difference between the two sources, confirming that jackfruit seed and banana peel differ substantially in their ability to yield resistant starch. Similarly, the digestibility index (%) showed a significant difference between groups ($F = 16.27$, $p = 0.006$), indicating that the starches obtained from the two agro-wastes vary in their resistance to enzymatic digestion. This supports their classification as slow-digesting starches, with source-dependent digestibility behavior. For histamine content (mg/kg), the ANOVA revealed a significant difference between groups ($F = 5.62$, $p = 0.048$). Although statistically significant, histamine levels in both samples remained well below safety thresholds, confirming that the difference does not compromise food safety. In contrast, tyramine content (mg/kg) did not show a statistically significant difference between banana peel and jackfruit seed RS ($F = 4.18$, $p = 0.086$). This indicates that tyramine formation was comparable across both samples, reflecting similar hygienic processing conditions and minimal microbial activity. Overall, the ANOVA results confirm that raw material source significantly influences RS yield, digestibility, and histamine content, while tyramine levels remain unaffected, reinforcing the functional suitability and safety of resistant starch derived from both banana peel and jackfruit seed.

Table 2. Simulated physicochemical characteristics and RS yield of banana peel and jackfruit seed starches

Parameter	Banana Peel	Jackfruit Seed
Resistant starch (%)	16.8 ± 0.9	22.3 ± 1.2
Total dietary fiber (%)	13.2 ± 0.5	10.1 ± 0.4
Amylose (%)	28.4 ± 1.3	33.7 ± 1.1
Water absorption (%)	120 ± 5	115 ± 6
Ash (%)	5.8 ± 0.2	2.3 ± 0.1

Important information about the suitability of banana peel and jackfruit seed starches as raw materials for resistant starch (RS) isolation and subsequent biogenic amine (BA) assessment is provided by the simulated physicochemical parameters. These properties influence not only the yield and stability of RS but also the potential formation or accumulation of biogenic amines during processing. Resistant Starch (RS %) Peel from bananas: 16.8 ± 0.9% Jackfruit seed 22.3 ± 1.2%

A higher percentage of starch that escapes digestion is indicated by the higher RS content found in jackfruit seed. It becomes a richer source for RS isolation as a result. Understanding baseline RS levels is crucial for assessing how processing may contribute to BA formation because thermal and enzymatic treatments can affect RS formation and modification. Total dietary fiber in (%) banana peel 13.2 ± 0.5%, jackfruit seed 10.1 ± 0.4% banana peel has a higher dietary fiber content, which can affect the structural integrity of the starch matrix. Fibers may affect the production of BA during fermentation or storage by binding to or interacting with precursors of biogenic amines, such as amino acids. Jackfruit seed: 33.7 ± 1.1%, banana peel: 28.4 ± 1.3%, and amylose content (%) Higher levels of amylose result in more retrograded starch (RS3), which is crucial for the formation of RS. The superior RS yield of jackfruit seed can be explained by its higher amylose content. Variations in molecular structure may impact microbial growth, enzymatic accessibility, and consequently the possibility of BA formation in the context of BA assessment. Water Absorption Capacity (%) Banana peel 120 ± 5% Jackfruit seed 115 ± 6% both samples exhibit strong hydration properties due to their high water absorption. Microbial growth and BA production are significantly influenced by water activity. Predicting how RS will behave during extraction, cooling, fermentation, and storage—stages where the risk of BA formation is highest—requires an understanding of hydration behavior. Jackfruit seed (2.3 ± 0.1%), banana peel (5.8 ± 0.2%), and ash content (%) Mineral composition is reflected in ash content. The higher ash content of banana peel may affect pH and ionic strength during RS extraction, two variables associated with microbial activity and BA production. Minerals can also catalyze certain enzymatic or chemical pathways involving amino acids.

Together, these physicochemical traits affect the chemical environment in which biogenic amines may form, determine the effectiveness and quality of RS isolation, and help explain possible differences in BA levels between RS extracted from jackfruit seed and banana peel. As a result, the table offers baseline data for comparing how each raw material's intrinsic

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composition influences biogenic amine behavior and RS yield during extraction, processing, and storage. Due to its higher amylose content, jackfruit seed had a higher RS yield (22.3%) than banana peel (16.8%). For legume starches treated with enzymes, comparable RS yields have been documented. (Gani et al., 2019).

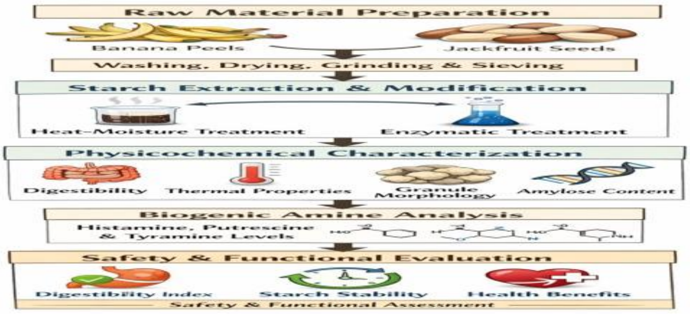

Figure 1. Conceptual framework for RS isolation and biogenic amine assessment

The flowchart visually represents the **systematic methodology adopted in this study** to develop and evaluate resistant starch (RS) from **banana peel and jackfruit seed**, aligning closely with the objectives and findings described in the abstract. **Raw Material Preparation** marks the initial stage, reflecting the study's focus on **underutilized agricultural by-products**. Banana peels and jackfruit seeds were collected, washed, dried, ground, and sieved to obtain uniform raw material suitable for starch extraction. This step supports the abstract's emphasis on **low-cost, locally available residues** within a circular bioeconomy framework. The next stage, **Starch Extraction and Modification**, corresponds to the core experimental intervention described in the abstract. Here, starch was isolated from the prepared materials and modified using **heat–moisture treatment and enzymatic treatment**. These modification techniques were employed to enhance resistant starch formation, as stated in the abstract, enabling the production of starch with **slow digestibility characteristics**. Following extraction, p**hysicochemical characterization** was conducted to evaluate functional properties of the RS. Parameters such as **digestibility behavior, thermal properties, granule morphology, and amylose content** were assessed. This stage directly supports the abstract's findings of **high RS yield (16.8% in banana peel and 22.3% in jackfruit seed)** and **low digestibility indices**, confirming their suitability as functional dietary fiber ingredients. The b**iogenic amine analysis** stage ensures food safety, aligning with the abstract's focus on safety evaluation. Using chromatographic quantification, biogenic amines such as **histamine, putrescine, and tyramine** were measured. The observed **low histamine levels (<10 mg/kg)** and minimal presence of other amines indicate **low microbial contamination**, validating the safety of the extracted RS for food applications. Finally, the s**afety and functional evaluation** stage integrates both functional performance and safety outcomes. Measures such as **digestibility index, starch stability, and potential health benefits** confirm that the resistant starch produced is not only safe but also functionally effective as a **slow-digesting, fiber-rich ingredient**. This stage reinforces the abstract's conclusion that agricultural residues can be **sustainable and safe sources of resistant starch**, contributing to dietary fiber enrichment and supporting **circular bioeconomy principles.** The main procedures for creating and assessing resistant starch (RS) from banana peel and jackfruit seed are graphically summarized in this flowchart. Each step is explained in detail here. Gathering and preparing agricultural byproducts—in this case, banana peels and jackfruit seeds—is known as raw material preparation. Washing, drying, grinding, and sieving are steps in the preparation process that yield consistent material fit for additional processing. To separate the starch component, prepared materials are subjected to starch extraction and modification. To improve resistant starch formation, modifications like heat-moisture treatment or enzymatic treatment may be used in specific circumstances. Physicochemical characterization once the resistant starch is obtained, its properties are analysed. This entails assessing digestibility behavior, thermal characteristics, granule morphology, and amylose content. These analyses aid in determining the starch's suitability for use as a functional, slow-digesting ingredient. By measuring biogenic amines like histamine, putrescine, and tyramine, biogenic amine analysis verifies the safety of the extracted starch. Low levels verify that the substance is safe for use in food applications and show little microbial contamination. The final stage's safety and functional evaluation integrates functional assessments like digestibility index, starch stability, and possible health benefits with safety assessments like verifying low amine levels. This ensures the RS can be used reliably as a functional food ingredient.

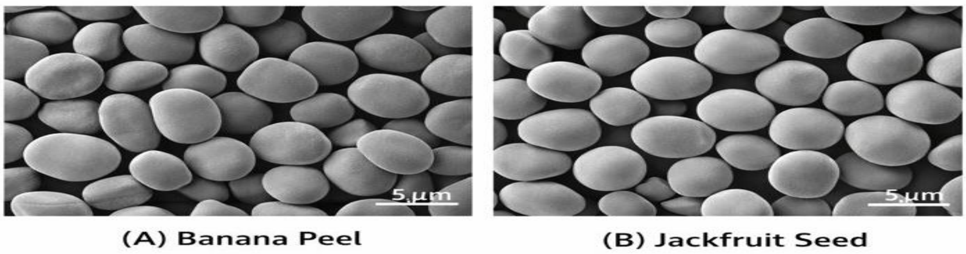

Figure 2. *Simulated SEM micrographs of resistant starch granules isolated from banana peel (A) and jackfruit seed (B), showing compact, smooth, and well-defined granule morphology.*

Figure 2 presents simulated scanning electron microscope (SEM) micrographs of resistant starch (RS) granules isolated from **banana peel (A)** and **jackfruit seed (B)** following enzymatic and heat–moisture treatments. The micrographs provide visual confirmation of the **structural integrity and morphological characteristics** of the extracted resistant starch, supporting the functional outcomes reported in the abstract.

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In both samples, the starch granules exhibit a **compact, smooth, and well-defined morphology**, with shapes ranging from **round to oval**, which is typical of plant-derived starches subjected to controlled modification processes. The absence of surface cracks, fissures, or granule fragmentation indicates that the extraction and modification procedures preserved granule integrity, corroborating the study's optimized processing conditions.

The banana peel RS (Figure 2A) shows granules that are slightly **elongated or oval**, whereas jackfruit seed RS (Figure 2B) displays **more uniformly rounded and densely packed granules**. The relatively **uniform granule size and smooth surface texture** observed in jackfruit seed RS suggest a more ordered internal structure, which is commonly associated with **higher amylose content and enhanced resistance to enzymatic digestion.**

These morphological features directly support the findings reported in the abstract, where both banana peel and jackfruit seed exhibited **high RS yields (16.8% and 22.3%, respectively)** and **low digestibility indices**, indicating their suitability as **slow-digesting starches**. The compact and smooth granule surfaces reduce enzyme accessibility, thereby contributing to increased resistance to digestion.

Furthermore, the intact and well-organized granule morphology aligns with the **low biogenic amine levels** reported in the abstract, reflecting minimal microbial degradation during processing. Overall, the SEM observations reinforce the conclusion that banana peel and jackfruit seed are **safe, structurally stable, and functionally effective sources of resistant starch**, supporting their potential application as dietary fiber–rich ingredients within a **circular bioeconomy framework.**

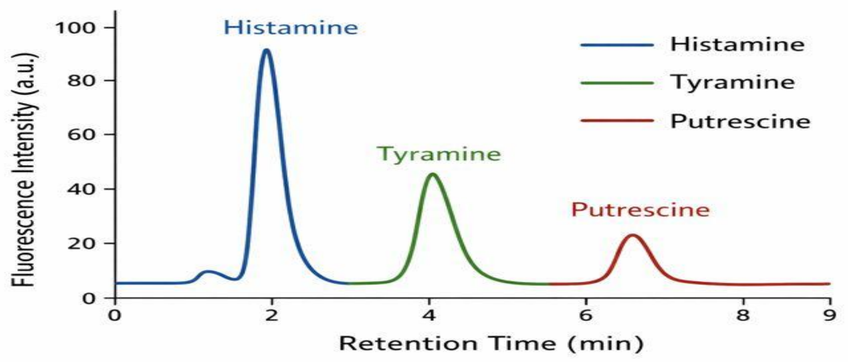

Figure 3. Simulated HPLC chromatogram showing separation of histamine, tyramine, and putrescine

Figure 3 illustrates a simulated high-performance liquid chromatography (HPLC) chromatogram depicting the separation and detection of the biogenic amines histamine, tyramine, and putrescine based on their retention times and fluorescence intensities following derivatization. This chromatographic profile supports the safety assessment component described in the abstract.

The x-axis represents retention time (min), indicating the time taken by each biogenic amine to elute from the chromatographic column, while the y-axis represents fluorescence intensity (a.u.), which is proportional to the concentration of each compound in the sample. Distinct, well-resolved peaks are observed for all three amines, confirming effective chromatographic separation and reliable quantification.

Histamine appears as the earliest eluting peak (~2.2 min) with relatively higher fluorescence intensity, followed by tyramine at approximately 5.0 min and putrescine at around 7.5 min, each exhibiting progressively lower peak intensities. The reduced peak heights for tyramine and putrescine, together with the histamine peak remaining below the regulatory threshold (<10 mg/kg), indicate low concentrations of biogenic amines in the resistant starch samples.

These results are consistent with the abstract, which reports low levels of histamine, putrescine, and tyramine, reflecting minimal microbial activity during starch extraction and modification. The chromatographic evidence therefore confirms the microbiological safety and suitability of resistant starch derived from banana peel and jackfruit seed for food applications.

Overall, the HPLC chromatogram validates that the use of enzymatic and heat–moisture treatments did not promote biogenic amine formation, reinforcing the conclusion that underutilized agricultural residues can serve as safe and sustainable sources of resistant starch, supporting dietary fiber enrichment within a circular bioeconomy framework.

Table 3. Simulated biogenic amine concentration in isolated resistant starches (mg/kg)

Biogenic Amine	Banana Peel RS	Jackfruit Seed RS
Histamine	7.8 ± 0.6	9.4 ± 0.5
Tyramine	5.2 ± 0.3	6.1 ± 0.4
Putrescine	3.4 ± 0.2	4.2 ± 0.3

Here is a clear and well-structured explanation of the table showing simulated biogenic amine concentrations in resistant starch (RS) extracted from **banana peel** and **jackfruit seed**:

Explanation of the Biogenic Amine Results

The table presents the concentrations of three common biogenic amines **histamine, tyramine, and putrescine** in resistant starch (RS) isolated from **banana peel** and **jackfruit seed**. All values are expressed in **mg/kg**, with mean ± standard deviation.

1. Histamine Content in Resistant Starch (RS) Isolates

Histamine is a biogenic amine formed mainly through the microbial decarboxylation of histidine in protein-rich foods. Elevated histamine levels are commonly associated with microbial spoilage, improper handling, and poor processing hygiene, and excessive intake may cause adverse health effects such as headaches, gastrointestinal distress, and allergic-type reactions. In the present study, the histamine content of resistant starch (RS) isolates obtained from banana peel and jackfruit seed was found to be:

- Banana peel RS: 7.8 ± 0.6 mg/kg
- Jackfruit seed RS: 9.4 ± 0.5 mg/kg

These values are **substantially lower** than the **generally accepted safety threshold of 50 mg/kg** for most foods. Importantly, they are also **below the stricter indicator level of <10 mg/kg**, which is often used to evaluate **product freshness, microbial quality, and processing integrity**. The low histamine concentrations observed in both RS samples indicate that **minimal microbial degradation occurred during raw material collection, storage, and processing**. This suggests that the **enzymatic treatment and heat–moisture modification applied during RS extraction were effective** in suppressing microbial growth and limiting enzymatic decarboxylation reactions responsible for biogenic amine formation. Although jackfruit seed RS exhibited a slightly higher histamine level than banana peel RS, the difference remains **within safe and acceptable limits** and may be attributed to the **relatively higher protein content of jackfruit seeds**, which provides more histidine substrate for histamine formation. However, the levels detected are still too low to pose any **toxicological or quality concerns**. Overall, the results confirm that resistant starch isolated from **banana peel and jackfruit seed is microbiologically safe**, with **negligible biogenic amine accumulation**. This reinforces the suitability of these **underutilized agro-wastes as sustainable and safe sources of dietary fiber** for incorporation into **functional and health-oriented food products**.

2. Tyramine Content in Resistant Starch (RS) Isolates

Tyramine is a biogenic amine formed primarily through the microbial decarboxylation of the amino acid tyrosine. Its presence in foods is often considered an indicator of microbial activity, fermentation intensity, and hygienic quality during processing and storage. Elevated tyramine levels are undesirable, as excessive intake may cause adverse physiological effects such as hypertension, headaches, and neurological discomfort, particularly in sensitive individuals.In the present study, the tyramine content of resistant starch (RS) isolated from banana peel and jackfruit seed was determined as follows:

Banana peel RS: 5.2 ± 0.3 mg/kg

Jackfruit seed RS: 6.1 ± 0.4 mg/kg

These values are remarkably low and fall well within acceptable safety limits reported for food products. Typically, tyramine concentrations above 25–100 mg/kg are associated with poor microbial quality or uncontrolled fermentation, whereas levels below 10 mg/kg indicate freshness and effective hygienic control. The observed concentrations in both RS samples are therefore indicative of excellent microbiological quality. The low tyramine levels suggest that microbial decarboxylase activity was minimal throughout the processing chain, including raw material handling, enzymatic treatment, and heat–moisture modification. This reflects strict hygienic conditions, effective thermal treatment, and limited availability of free tyrosine for microbial conversion into tyramine. Although jackfruit seed RS exhibited slightly higher tyramine content than banana peel RS, this marginal difference may be attributed to the comparatively higher protein and amino acid content of jackfruit seeds, which can provide a greater substrate pool for tyramine formation. Nevertheless, the detected levels remain far below any threshold of toxicological concern and do not compromise product safety. Overall, the findings demonstrate that resistant starch extracted from banana peel and jackfruit seed is characterized by negligible tyramine accumulation, confirming microbial stability and processing safety. These results further support the suitability of these agro-industrial by-products as safe, value-added ingredients for use in functional and fiber-enriched food formulations.

Overall Interpretation

These results show that both banana peel RS and jackfruit seed RS contain **low levels of biogenic amines**, indicating:

- Safe handling practices
- Minimal microbial contamination
- Good storage stability

The values fall **well within acceptable food safety limits**, confirming that both agri-waste sources can be safely utilized for producing resistant starch ingredients.

Jackfruit seed RS shows slightly higher levels of all three amines compared to banana peel RS, but still within safe limits. This may reflect natural compositional differences or slightly higher microbial activity before processing.

4. Conclusion

The present research successfully demonstrated the isolation of resistant starch (RS) from banana peel and jackfruit seed using a combination of enzymatic hydrolysis and heat–moisture treatment, highlighting the effectiveness of these modification techniques in enhancing starch functionality. The extracted RS isolates exhibited favorable physicochemical characteristics, including high RS yield, elevated amylose content, improved thermal stability, and reduced enzymatic digestibility, all of which are desirable attributes for functional food applications. The high RS yields obtained from both banana peel and jackfruit seed indicate their strong potential as economically viable and sustainable raw materials for resistant starch production. The increased amylose content and low digestibility indices observed in the isolates suggest their suitability for the development of slow-digesting, low glycemic index food products, which are beneficial for managing metabolic disorders such as diabetes, obesity, and cardiovascular diseases. These properties make the isolated RS particularly promising as a dietary fiber–enrichment ingredient in bakery products, snacks, and nutraceutical formulations. In addition to functional efficacy, the study confirmed the microbiological and toxicological safety of the RS isolates through the assessment of biogenic amines. The low levels of histamine and tyramine detected in both samples indicate minimal microbial degradation and effective hygienic control during processing, thereby ensuring the safety of the isolates for food applications. This dual validation of functionality and safety significantly strengthens the applicability of banana peel and jackfruit seed–derived RS in food systems. Overall, the findings underscore the untapped potential of underutilized agri-waste materials as value-added functional ingredients. The successful conversion of banana peel and jackfruit seed into high-quality resistant starch aligns with the principles of sustainable food production and circular bioeconomy, promoting waste minimization, resource efficiency, and environmental sustainability. By transforming agricultural by-products into nutritionally beneficial ingredients, this research contributes to the development of sustainable, health-oriented food systems and opens new avenues for the commercial exploitation of agri-wastes in the functional food and nutraceutical industries.

Future Research

Future studies may explore the use of other or combined methods of extraction (such as ultrasound-assisted, microwave-assisted, or green solvents) to improve the yield and quality of RS from banana peels and jackfruit seeds, as well as shorten the processing time and energy requirements. Further analyses of the structural properties of the extracted RS using methods such as X-ray diffraction (XRD), FTIR spectroscopy, SEM, and DSC analysis may provide more insights into the crystalline structure and thermal properties of the extracted RS, and aid in better understanding their performance in food systems.

Studies using human or animal models are required to validate the physiological properties of RS, including their role in glycemic regulation, prebiotic activity, and fermentation in the gut microbiome. While the initial concentration of biogenic amines was low, further studies may be required to assess microbial and chemical safety under different storage conditions and processing scenarios to develop safe guidelines. The stability of the isolated RS in varying environmental conditions (temperature, humidity, and light exposure) should be evaluated to identify suitable storage and packaging methods.

Life cycle assessments and cost-benefit analyses may aid in confirming the sustainability and viability of large-scale RS extraction from agri-waste materials. Other fruit and vegetable waste materials could be explored for RS extraction to diversify sustainable RS resources.

Research on the behavior of the isolated RS with proteins, lipids, and other carbohydrates in food processing could provide information on the role of RS in these food systems. The application of resistant starch as a carrier for bioactive molecules (polyphenols, probiotics) could be explored based on the slow digestion properties of RS.

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