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Resistant starch from yam (*Dioscorea alata* L.) as a potential prebiotic and microencapsulation matrix for enhancing the viability of *Lacticaseibacillus paracasei* D4

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ABSTRACT

Resistant starch (RS) derived from tubers is a functional food ingredient with potential as a prebiotic and a matrix for probiotic microencapsulation. However, the exploration of resistant starch from Indonesian tuber sources remains limited. This study aimed to characterize starch and resistant starch type-3 (RS3) from yam (*Dioscorea alata* L.), sweet potato (*Ipomoea batatas* L.), and taro (*Colocasia esculenta* L.), evaluate their prebiotic activity toward *Lacticaseibacillus paracasei* D4, and assess probiotic viability under simulated gastrointestinal conditions. Starch was extracted and physically modified through autoclaving–cooling cycles to produce RS3. Prebiotic activity was evaluated using the prebiotic activity score (PAS) and the prebiotic index (PI). Microencapsulation was prepared by extrusion using a sodium alginate–chitosan–RS matrix, and viability was assessed through a three-phase gastrointestinal simulation (GIS). Yam starch (YS) showed the highest amylose (41.92 ± 0.77%) and RS content (6.12 ± 0.03%), as well as the highest prebiotic activity at a 2% concentration (PAS: 7.58 ± 4.97; PI: 1.19 ± 0.04). Yam RS microcapsule (YRSM) demonstrated the lowest viability decrease during GIS (1.45 log CFU/g), compared to the control microcapsule (3.15 log CFU/g). Scanning electron microscopy (SEM) analysis confirmed that YRSM maintained the best structural integrity post-GIS, as evidenced by minimal pore formation. These findings position YRS as an effective prebiotic and encapsulation matrix for synbiotic functional food development.

Contribution/Originality: This study is one of the very few studies that have investigated resistant starch (RS3) from local Indonesian tubers (sweet potato, taro, and yam) for dual-function properties as prebiotics and microencapsulation matrices for *Lacticaseibacillus paracasei* D4. The paper's primary contribution is finding that yam-derived RS3 was most effective in maintaining probiotic viability during simulated gastrointestinal digestion.

1. INTRODUCTION

Probiotics are defined as non-pathogenic microorganisms that confer health benefits when administered in adequate amounts (at least 10^6–10^7 CFU/g) (da Silva, Tagliapietra, do Amaral Flores, & dos Santos Richards, 2021; Hill et al., 2014; Latif et al., 2023). *Lacticaseibacillus paracasei* D4 is a lactic acid bacterium isolated from fermented coffee (wine coffee) that has been characterized to evaluate its potential as a probiotic (Jatmiko et al., 2025). However,

the viability of probiotics decreases in the gastrointestinal tract due to gastric acid, bile salts, and the activity of enzymes in the small intestine (Torp, Bahl, Boisen, & Licht, 2022). The microencapsulation technique has been developed to address this issue (Nguon, Lagugné-Labarthet, Brandys, Li, & Gillies, 2018; Sbehat, Mauriello, & Altamimi, 2022). Sodium alginate serves as the most widely used microencapsulation matrix because it is biocompatible and non-toxic to cells, but it is sensitive to extreme pH levels and produces a porous gel (Etchepare et al., 2016; Etchepare et al., 2015; Goh, Heng, & Chan, 2012). The stability of alginate-based microcapsules can be improved through chitosan coating (Zanjani, Tarzi, Sharifan, & Mohammadi, 2014) and the addition of prebiotics as components of a synbiotic matrix (Goderska & Kozłowski, 2021; Jagtiani & Adsare, 2022). Resistant starch (RS) is a potential prebiotic for use as a microencapsulation matrix (Muhammad, Ramzan, Zhang, & Zhang, 2021) as it is not subject to hydrolysis by stomach acid or digestive enzymes (Setiarto, Kusumaningrum, Jenie, Khusniati, & Sulistiani, 2020). Based on their mechanisms of resistance to human digestion, resistant starches are classified into several types, including type 3 (RS3) (Bojarczuk, Skąpska, Khaneghah, & Marszałek, 2022). Starches with higher amylose content tend to yield higher RS3 through retrogradation (Feng et al., 2024).

In the field of microencapsulation, the addition of RS can improve encapsulation efficiency and maintain probiotic viability during gastrointestinal tract simulation (Zanjani et al., 2014). The increased encapsulation efficiency is related to the characteristics of RS granules, which can reduce porosity and slow the diffusion of acid and bile salts (Jan et al., 2025). RS2 from Hi-Maze 260, as a prebiotic, has been reported to increase the population densities of *Bifidobacterium* and *Lactobacillus* and to inhibit the growth of enteric pathogenic bacteria (*Escherichia-Shigella*) in an *in vitro* fermentation model simulating the human gut microbiota (Zheng et al., 2025). Meanwhile, RS from green bananas, used as an encapsulation matrix, has been reported to protect the viability of a probiotic cocktail (*B. longum*, *Lactobacillus casei*, *L. rhamnosus*, *Streptococcus thermophilus*, and *Clostridium butyricum*) against acid stress (pH 2), bile salts, and simulated intestinal conditions (Wang, Yeh, Huang, Wu, & Chen, 2024). However, the protective effect of RS as an encapsulation matrix varies among starches; a previous study reported that the addition of Hi-Maze only improved encapsulation efficiency but failed to maintain probiotic viability under conditions of acid and bile salt stress (Sultana et al., 2000). The stability of different RS microencapsulation matrices is closely related to the amylose content of the starch, the type of RS, the RS preparation method, and the matrix formulation combination used. Previous research by Luca and Oroian (2021) compared different microencapsulation matrix sources, such as potato starch, inulin, oligofructose, and glucose, which yielded varying probiotic viability responses under simulated gastrointestinal conditions. These findings indicate that the physicochemical characteristics of the microencapsulation matrix influence its protective efficacy in maintaining probiotic viability.

Indonesia has a wide variety of local tuber crops with potential as starch sources (Aprianita, Vasiljevic, Bannikova, & Kasapis, 2014), including '*gembili*' (*Dioscorea esculenta* L.), '*ganyong*' (*Canna edulis* Kerr), '*beneng*' (*Xanthosoma undipes* K. Koch), taro (*Colocasia esculenta* L.), sweet potato (*Ipomoea batatas* L.), and yam (*Dioscorea alata* L.) (Aprianita et al., 2014; Horison & Surini, 2024; Lopulalan, Marseno, Marsono, & Pranoto, 2023). However, their utilization, especially in the case of taro and yam, remains limited. Additionally, these tubers are classified as minor crops with low commodity priority in Indonesia (Hapsari, Mas'udah, & Fauziah, 2023; Maharani & Arzani, 2024; Maretta, Sobir, Helianti, Purwono, & Santosa, 2021; Walsen, Polnaya, Lesilolo, Rehatta, & Lawalata, 2020). Sweet potato is a potential starch source, but its functional properties remain underexplored (Cui, Mahfouzi, Zhang, Gao, & Guo, 2024). Previous studies have separately reported the dual function of RS. RS from purple yam (*D. alata* L.) has been reported to act as a prebiotic that enhances the tolerance and proliferation of *Bifidobacterium adolescentis* under simulated gastrointestinal stress conditions (Li, Chen, et al., 2018). Another study by Erickson, Carlson, Stewart, and Slavin (2018) reported that RS from different sources, such as potatoes, tapioca, and corn, exhibited varying prebiotic activity even when tested under the same fermentation conditions. This indicates that the chemical properties of starch positively influence its prebiotic activity. Previous studies have reported only the functional properties of RS as a fermentation substrate, without further examining its potential as a microencapsulation matrix; thus, the relationship

between RS's prebiotic activity in promoting the growth of probiotics and its protective capacity as a microencapsulation matrix has not been extensively studied.

Furthermore, taro, yam, and sweet potatoes as local tubers from Indonesia have high amylose content (Boahemaa, Dzandu, Amissah, Akonor, & Saalia, 2024; Lyu et al., 2021; Zang, Gong, Cao, Ni, & Chang, 2024), which have the potential to yield RS3. Therefore, further research is needed to evaluate the dual function of RS as both a prebiotic and a microencapsulation matrix. The differences in the chemical characteristics of taro, sweet potato, and yam are thought to result in varying levels of RS3, which may influence their prebiotic activity and protective effects as a microencapsulation matrix. Therefore, this study aimed to (i) analyze the chemical characteristics of starch and resistant starch in yam, taro, and sweet potato, (ii) analyze the potential of the best resistant starch as a prebiotic to support the growth of *L. paracasei* D4, and (iii) analyze the potential of the best resistant starch as a microencapsulation matrix to maintain the viability of *L. paracasei* D4 during simulated gastrointestinal transit.

2. MATERIALS AND METHODS

2.1. Starch Extraction and Purification

Starch was extracted from taro (*C. esculenta* L.), sweet potato (*I. batatas* L.), and yam (*D. alata* L.) obtained from a local market in Malang, Indonesia. The starch extraction and purification processes followed the method described by Simsek and El (2012) with some modifications. Peeled and chopped tubers were blended with water (1:1, w/v) using a blender (Philips, HR2115/01). The mixture was filtered through a piece of double-layered filter cloth and centrifuged at 4,000 g for 20 minutes. The crude starch obtained was then gradually purified through an alkali treatment (0.05 M NaOH, 1:3, w/v; 6,000 g, 15 minutes), acid washing (0.1 M HCl), and defatting using chloroform and methanol (2:1, v/v), then dried at 50°C for 12 hours and stored at room temperature.

2.2. Production of Resistant Starch

The preparation of RS3 was based on the method described by Simsek and El (2012) with some modifications. A starch suspension (8%) was heated in a boiling water bath (BENCHMARK, SB-12L) for 10 min with stirring. The starch gel was gelatinized using an autoclave (TOMY, ES-215) at 121°C for 15 min. The gelatinized starch was stored in a refrigerator at 4°C (SHARP, SJP612NLVSL) overnight for retrogradation. The heating, autoclaving, and cooling processes were repeated once more to increase the RS3 content. The retrograded starch was dried at 60°C for 8 hours. The resulting resistant starch was stored in a tightly sealed, dry container at room temperature.

2.3. Chemical Characteristics Analysis of Starch and Resistant Starch

The chemical characteristics of starch from local tubers were analyzed, including amylose content, total starch content, and resistant starch content. Meanwhile, RS3 was analyzed only for its resistant starch content. Each parameter was evaluated using a completely randomized design (CRD). Each analysis was conducted using three replicates of starch samples for each tuber type. Amylose content was determined following AOAC International (2005). A mixture of amylose and amylopectin (0, 10, 25, 50, and 75% amylose) was used as a standard solution to prepare a standard curve for determining amylose content. Total starch was measured using AOAC International (2005). Glucose solutions (0, 10, 25, 50, and 75%) were used as standard solutions to prepare a calibration curve as a reference for determining total starch content by spectrophotometry. Total starch content was calculated using Formula 1. Resistant starch content was measured using the method described by Englyst, Kingman, and Cummings (1992) and calculated using Formula 2.

$$\text{Total starch (\%)} = A \times F \times V \times \frac{1}{1000} \times \frac{1}{W} \times \frac{162}{180} \times 1000 \quad (1)$$

Where A is the absorbance of the sample solution, F is the conversion factor of absorbance to glucose concentration (mg/mL), V is the volume correction factor, and W is the sample weight.

$$\text{Resistant starch content (\%)} = (\text{total starch} - \text{digestible starch}) \times 100\% \quad (2)$$

2.4. Prebiotic Activity Assay

Prebiotic activity was evaluated using 10% buffered peptone water (BPW) supplemented with RS as the growth medium. BPW was used to replace De Man, Rogosa, and Sharpe (MRS) broth and nutrient broth (NB), which are commonly used as bacterial growth media in prebiotic activity assays. This modification was based on preliminary test results showing that the use of MRS broth, NB, or 100% BPW as growth media resulted in low prebiotic efficacy for RS and that RS as a prebiotic could not be selectively utilized by probiotics alone. The BPW concentration was reduced to 10% to lower the nutrient content of the fermentation medium, thereby making RS3 the primary carbon source utilized during fermentation. Consequently, the prebiotic effect of RS3 on bacterial growth can be observed more specifically and accurately.

The prebiotic activity assay was modified from the method described by Kustyawati, Nurlita, Fadhallah, and Rizal (2022). The experiment was conducted using a two-factor factorial CRD. Each treatment combination (resistant starch type and concentration) was analyzed in three fermentation medium bottles. The bacteria used in this experiment were *L. paracasei* D4 and *Escherichia coli* ATCC 25922 (enteric pathogenic bacteria). *L. paracasei* D4 was obtained from the collection of the Microbiology Laboratory, Universitas Brawijaya. A bacterial inoculum was inoculated at a cell density of 10^6 cells/mL into a sterile fermentation medium (1:100, v/v), and the bacteria were fermented at 37°C for 24 h. The fermentation medium consisted of 5 mL of 10% buffer peptone water (BPW) with 1% glucose as a positive fermentable-carbon control and 5 mL of 10% BPW with RS3 at concentrations of 0.5%, 1%, and 2% as prebiotics. The concentrations of RS3 were selected based on (Rattanakiat, Pulbutr, Khunawattanakul, Sungthong, & Saramunee, 2020). The fermentation medium was sterilized at 121°C and 1 atm for 15 min. The total number of bacteria was counted at 0 and 24 hours of fermentation. Prebiotic activity was analyzed using the prebiotic activity score (PAS) and the prebiotic index (PI). The prebiotic activity score was calculated using Formula 3 (Rana et al., 2024), while the prebiotic index was determined using Formula 4 (Kustyawati et al., 2022).

$$\text{PAS} = \frac{(\text{LogP24}-\text{LogP0})\text{prebiotic}-(\text{LogE24}-\text{LogE0})\text{prebiotic}}{(\text{LogP24}-\text{LogP0})\text{glucose}-(\text{LogE24}-\text{LogE0})\text{glucose}} \quad (3)$$

Where P24 is the cell density of *L. paracasei* D4 at 24 h, P0 is the cell density of *L. paracasei* D4 at 0 h, E24 is the cell density of *E. coli* ATCC 25922 at 24 h, and E0 is the cell density of *E. coli* ATCC 25922 at 0 h.

$$\text{PI} = \frac{\text{cell density of } \textit{L. paracasei} \text{ D4 in prebiotic (CFU/mL)}}{\text{cell density of } \textit{L. paracasei} \text{ D4 in glucose (CFU/mL)}} \quad (4)$$

2.5. Production of Probiotic Microencapsulation

Probiotic microencapsulation was prepared using the extrusion method described by Rifa'i et al. (2025). A sterile mixture of 1.5% sodium alginate (Merck, 71238-50G) and resistant starch was prepared as the microencapsulation matrix. The concentration of resistant starch in each tuber was determined from the prebiotic activity assay results. For the negative control group, only sodium alginate was used as the matrix, without the addition of resistant starch. A culture of *L. paracasei* D4 at a density of 10^{10} cells/mL was inoculated into the sterile matrix mixture (1:1, v/v). The mixture was extruded through a 22G needle into 0.1 M CaCl2. The microcapsules were soaked in 0.5% sterile chitosan (Merck, 44887-50G) for 40 minutes and then washed and stored in a 0.1% peptone solution 4°C.

2.6. Viable Cell Count of Microencapsulated Probiotics

The viable cell count of microencapsulated probiotics was determined according to the method described by Moghanjougi, Bari, Khaledabad, Amiri, and Almasi (2021), with modifications. One gram of microcapsules was inoculated into 9 mL of sterile 0.1 M sodium citrate solution (pH 7) and then vortexed for 10 minutes. The bacterial suspension was diluted into a 0.85% NaCl solution (1:9, v/v) using the serial dilution method, then inoculated into MRS agar medium using the pour plate method and incubated at 37°C for 48 hours. The cell count was calculated using the total plate count.

2.7. Gastrointestinal Simulation Assay

Gastrointestinal simulation was based on the method described by da Silva et al. (2021), which consists of the stomach, duodenum, and ileum phases. The tests were evaluated using CRD, with three replicates of the microcapsule samples per treatment group. In the gastric phase, one gram of microcapsules was inoculated into 9 mL of BPW. To the same aliquot, 0.05 mL of 25 mg/mL pepsin was added gradually in six steps (pH/t: 5.5/10 min, 4.6/10 min, 3.8/10 min, 2.8/20 min, 2.3/20 min, and 2.0/20 min) and incubated in a rotary incubator at 37°C and 130 rpm. The simulation continued to the duodenum phase; the microcapsules were inoculated into 9 mL of 0.1% BPW, with 0.125 mL of 2 g/L pancreatin and 0.125 mL of 12 g/L bovine bile salt. The sample was adjusted to pH 5 and incubated at 37°C and 45 rpm for 20 min. The simulation continued to the ileum phase; the microcapsules were inoculated into the same mixture as in the duodenum simulation, but the pH was adjusted to 6.5, and incubation was conducted at 37°C and 45 rpm for 90 min. Probiotic viability in the stomach, duodenum, and ileum phases was calculated according to Moghanjougi et al. (2021).

2.8. Scanning Electron Microscopy of Microcapsules

The surface morphology of the microcapsules was observed using a scanning electron microscope (SEM). The microcapsules were observed under two conditions, pre- and post-gastrointestinal simulations, at the Integrated Research Laboratory of Universitas Brawijaya. The microcapsules were vacuumed at high power and then coated with gold for electron microscopy. Afterward, the microcapsules were observed using a TM-3000 electron microscope equipped with a SwiftED-3000 X-ray Microanalyzer (Hitachi, Japan), operated at 15 kV. SEM images were analyzed using ImageJ 1.54 software to determine changes in the pores on the surfaces of the microcapsules. SEM images were analyzed using ImageJ software to determine changes in pore size and pore count on the microcapsule surface. Pore segmentation was performed using pixel-intensity thresholding and the watershed method to separate adjacent pores. Pores were identified using the "Analyze Particles" feature with a minimum size threshold of 100 pixels^2. Pore size was reported as Feret's diameter (μm), and the number of pores was obtained from the pores detected on the overlay mask.

2.9. Statistical Analysis

Data are expressed as means ± standard deviations. Data on the chemical characteristics of starch and resistant starch, pore size, and those on probiotic viability derived during the gastrointestinal tract simulation were analyzed using one-way analysis of variance (ANOVA), while data from the prebiotic activity test were analyzed using two-way ANOVA. Comparisons between treatments were tested using Tukey's Honestly Significant Difference (HSD) test with SPSS software version 24.0. Differences were considered statistically significant at $p < 0.05$. Each test was conducted in triplicate to ensure the reliability and reproducibility of the data.

3. RESULTS AND DISCUSSION

3.1. Yield and Compositional Properties of Starch and Resistant Starches from Local Tubers

Starch extraction from three types of local tubers showed significant differences in yield ($p < 0.05$). Yam starch (YS) extraction showed the highest yield of 21.40 ± 2.15%, followed by sweet potato starch (SPS) at 15.20 ± 1.23%. Meanwhile, the lowest yield was from taro starch (TS) at 11.70 ± 0.67% (Table 1). These differences were closely related to the granule size of each starch; TS had small granules (1–5 μm) (Agama-Acevedo et al., 2011), causing it to settle more slowly and tend to become trapped within the fiber matrix during extraction (Zhu, 2015). YS had medium-to-large granule sizes (18–50 μm), and SPS had small-to-medium granule sizes (2.00–25.86 μm) (Jayakody, Hoover, Liu, & Donner, 2007; Tanimola, Otegbayo, & Akinoso, 2022). Other factors that could influence yield differences among tubers included harvest time, storage period, and cultivation conditions (Silva et al., 2020).

Table 1. Chemical characteristics of starch and resistant starch in local tubers.

Source	Yield (%)	Amylose (%)	Amylopectin (%)	Total starch (%)	Resistant Starch (%)
Starch					
Taro	11.17 ± 3.42^{a}	20.01 ± 0.40^{a}	51.77 ± 3.08^{c}	71.78 ± 2.68^{a}	2.56 ± 0.04^{a}
Sweet potato	15.20 ± 1.23^{b}	33.03 ± 0.43^{b}	42.97 ± 0.67^{b}	76.00 ± 0.36^{b}	3.81 ± 0.04^{b}
Yam	21.40 ± 2.15^{c}	41.92 ± 0.77^{c}	37.96 ± 2.89^{a}	79.88 ± 2.12^{c}	4.01 ± 0.03^{c}
Resistant starch					
Taro	24.22 ± 3.42^{A}	-	-	-	5.55 ± 0.04^{d}
Sweet potato	32.22 ± 2.69^{B}	-	-	-	5.85 ± 0.06^{e}
Yam	38.89 ± 2.34^{C}	-	-	-	6.12 ± 0.03^{f}

Note: Data are presented as means ± standard deviations of triplicate measurements. ^{abcdef}different lowercase superscript letters within the same column indicate significant differences among treatments for starch characteristics based on Tukey's HSD test ($p < 0.05$). ^{abc} different uppercase superscripts within the same column indicate significant differences among treatments for resistant starch yield based on Tukey's HSD test ($p < 0.05$).

The total starch content of YS (79.88 ± 2.12%) was significantly different from that of TS (71.78 ± 2.68%) (Table 1). Yam starch had the highest starch content among SPS and TS. The high total starch content in YS indicated a higher purity level of the extract. In turn, a high purity level represented lower ash, protein, and lipid content (Andrade, Barbosa, & Pereira, 2017). In different cultivars, previous studies reported YS content similar to these findings, such as Kahata Ala (74.11 ± 0.27%) and Hingurala (69.41 ± 0.54%) (Chiranthika, Chandrasekara, & Gunathilake, 2022). Other studies have reported lower total starch content in YS than in this study, such as those by Ochoa and Osorio-Tobón (2024) (41.23%) and Afoakwa, Polycarp, Budu, Mensah-Brown, and Otoo (2013) (63.24%). Meanwhile, it was reported that the São Tomé cultivar of YS had a higher total starch content than that in this study (96.88 ± 0.18%). Meanwhile, other cultivars showed a wide variation (41.23–96.88%) (Englyst et al., 1992; Kustyawati et al., 2022; Rattanakiat et al., 2020). Variations in total starch content among yam tubers were influenced by plant genetic factors and environmental conditions (temperature and light intensity) (Rosida, Harijono, Teti, & Endang, 2016). High total starch content in yam implies better gelatinization quality during heating, as more starch is available to form a dense and strong gel matrix (Adesokan et al., 2025).

The amylose content of YS showed a significant difference from that of TS. YS had the highest amylose content (41.92 ± 0.77%), followed by SPS (33.03 ± 0.43%), while TS had the lowest amylose content (20.01 ± 0.40%). Meanwhile, amylopectin content showed a different trend, with the highest content in TS (51.77 ± 3.08%) and the lowest in YS (37.96 ± 2.89%) (Table 1). The amylose-to-amylopectin ratio is a primary chemical parameter representing the functional properties of starch, including gel-forming ability, viscosity, and retrogradation tendency Cornejo-Ramírez et al. (2018). Zang et al. (2024) also reported a high amylose content in YS. Amylose content varied among different yam varieties, ranging from 4.83% to 28.70%. These findings are similar to those of Ndisya, Mbuge, Gitau, and Sturm (2026), who reported that YS and SPS had higher amylose content compared to TS, resulting in a higher amylopectin content in taro starch than in amylose. The ratio of amylose to amylopectin in these tuber starches was influenced by growing environmental conditions and botanical factors.

Native RS content differed significantly among the three types of starch ($p < 0.05$), with the highest RS content found in yam starch (4.01 ± 0.03%). YS had the highest RS content, followed by SPS (3.81 ± 0.04%) and TS (2.56 ± 0.04%) (Table 1). The differences in RS content among species were influenced by several factors, such as the chemical and physical characteristics of starch granules, starch crystallinity patterns, and molecular interactions and structures (Thuy et al., 2022). In foods, differences in RS content are primarily influenced by the amylose-to-amylopectin ratio (Zhao, Andersson, & Andersson, 2018). Previous studies by Aprianita et al. (2014) reported similar findings, where YS had a higher RS content (13.20 ± 0.80%) than SPS (10.20 ± 0.40%) and TS (3.3 ± 1.20%). Several different varieties of yam (*D. alata* L.) showed higher RS content than in this study, including Da Shan Line 2 (9.20%), Tainung 5 (10.80%), and Heng Chun (9.20%) (Li & Chou, 2018), as well as Kahata Ala (7.52%) and Hingurala (4.65%) (Chiranthika et al., 2022). Sweet potato (*I. batatas* L.) starch exhibited varying RS contents in several varieties, ranging from 1.76% to 27.75% (Kim et al., 2020).

The physical modification process through two cycles of autoclaving and cooling significantly increased the RS (RS3) content in all three types of tubers. Yam resistant starch (YRS) showed the highest RS3 yield (38.89 ± 2.34%), followed by sweet potato resistant starch (SPRS) (32.22 ± 2.69%) and taro resistant starch (TRS) (24.22 ± 3.42%), with RS3 contents of 6.12 ± 0.03%, 5.85 ± 0.06%, and 5.55 ± 0.04%, respectively (Table 1). RS content increased through the gelatinization of starch granules during autoclaving, followed by amylose retrogradation during cooling, which formed a new crystalline structure more resistant to enzymatic hydrolysis (Soral-Śmietana, Wronkowska, Biedrzycka, Bielecka, & Ocicka, 2005). These findings were similar to those of Rosida et al. (2016), who reported an increase in RS content from 2.22% to 9.04% after physical modification of the starch.

3.2. Prebiotic Activity of Resistant Starch

Based on Figure 1, YRS showed an increasing trend in PAS with increasing concentration, with the 2% concentration showing the highest average PAS of 7.58 ± 4.97. Meanwhile, SPRS showed a decrease in PAS as the concentration increased. The decrease in PAS as the concentration increased may have been due to several factors. The first factor was substrate inhibition: too high a substrate concentration actually inhibited the growth of probiotic bacteria (Zheng & Mao, 2026). Another contributing factor was that an excessively rapid fermentation rate at high substrate concentrations may have triggered an extreme drop in pH, which, in turn, created a feedback inhibition effect on probiotic population growth (Sarvari, Mortazavian, & Fazeli, 2014). Different from YRS and SPRS, TRS showed negative PAS values at the 0.5% and 1% concentrations, indicating that TRS was not only selectively metabolized by probiotics but could also be metabolized by enteric pathogenic bacteria. Statistical analysis showed no significant difference among the three types of RS in prebiotic activity against *L. paracasei* D4 ($p = 0.224$). However, the concentration with the highest score for each type of RS was still used as the basis for preparing microcapsules.

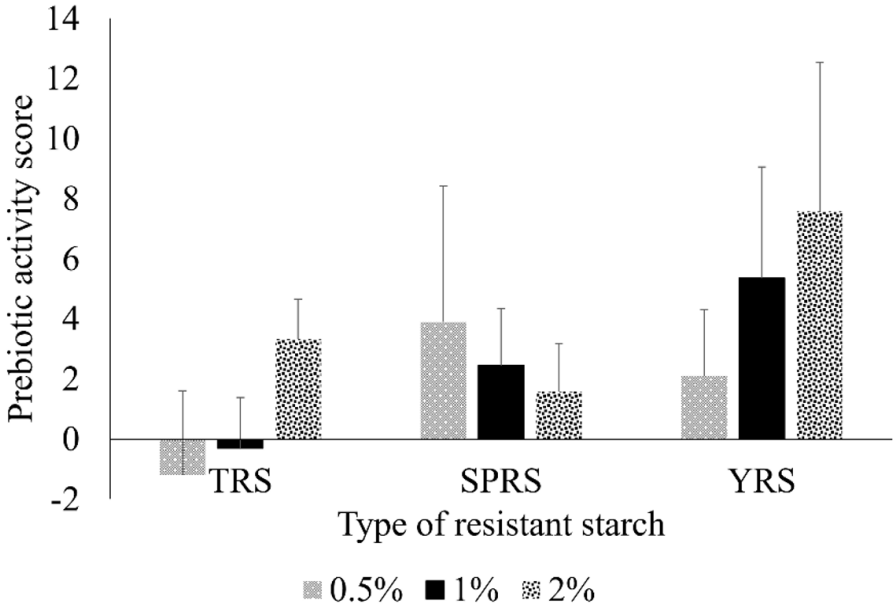

Figure 1. Prebiotic activity scores of resistant starch from local tubers.

Note: Mean values are not significantly different according to Tukey's HSD test ($p > 0.05$). TRS = Taro resistant starch; SPRS = Sweet potato resistant starch; YRS = Yam resistant starch.

The variations in PAS among resistant starches were similar to those found in previous studies, which reported that metabolic diversity in Lactobacilli caused variations in PAS among different types of prebiotics metabolized by the same probiotic strain (Figueroa-González, Rodríguez-Serrano, Gómez-Ruiz, García-Garibay, & Cruz-Guerrero, 2019; Kustyawati et al., 2022). The variations in PAS among the same probiotic strain also aligned with previous research; Lee, Lee, Kim, Shin, and Park (2023) reported that *L. paracasei* ATCC 27092 showed a PAS value of less than 1.3 when fermenting 0.5% green radish (*Raphanus sativus* L.) polysaccharides and a negative PAS value when

fermenting fructooligosaccharides (FOS) (-0.051) (Silva, Garcia, Baldo, & Celligoi, 2017). Another previous study by Palacio, Etcheverría, and Manrique (2014) reported that *L. paracasei* BGP1 also exhibited varying PAS values, including in raffinose (0.37 ± 0.05), yellow squash (*Cucurbita maxima*) (0.55 ± 0.03), and lupin seed extracts (*Lupinus albus*) (0.49 ± 0.02).

Previous studies have reported the prebiotic activity of yam, taro, and sweet potato using different processing methods. Buenaventura et al. (2025) reported that 1% purple yam jam (*D. alata* L.) showed a lower PAS value than 2% YRS, which was 0.77. Sweet potato flour at 20 g/L from four different varieties also showed lower PAS values than 0.5% SPRS, including Rainha Branca (0.18 ± 0.03), Campina Branca (0.28 ± 0.01), Vitória (0.32 ± 0.04), and Lagoinha (0.40 ± 0.05) (de Albuquerque et al., 2020). TRS from several physical modification processes also showed negative results, similar to PAS values for 0.5% and 1% TRS, such as RS subjected to three cycles of autoclaving–cooling (-0.372), annealing (-0.211), and heat-moisture treatment (-0.342). However, TRS subjected to two cycles of autoclaving–cooling showed a positive PAS result of 0.072 (Setiarto et al., 2019). Inulin, as a commercial prebiotic, showed a lower PAS value when fermented by *L. paracasei* 1195 than 2% YRS, which was 1.17 (Huebner, Wehling, & Hutkins, 2007); this indicates that YRS had greater potential for stimulating probiotic growth.

The prebiotic index (PI) is a quantitative method used to assess the effectiveness of a substrate in supporting probiotic growth. A PI value greater than 1 ($x > 1$) indicates that the prebiotic has a positive effect on probiotic growth. In contrast, a PI value less than 1 ($x < 1$) indicates that the prebiotic has low effectiveness in supporting probiotic growth (Figueroa-González et al., 2019). YRS showed the highest PI value at 2% (1.19 ± 0.04) (Figure 2) and exhibited an increase in PI as its concentration increased, while SPRS showed a decrease in PI as its concentration increased. At each concentration, both YRS and SPRS showed PI values > 1, indicating that both types of RS had a positive effect as prebiotics in supporting the growth of *L. paracasei* D4. The PI values for TRS at the concentrations of 0.5% and 1% were less than 1 (PI < 1), indicating that, at these concentrations, TRS had low effectiveness as a prebiotic to support the growth of *L. paracasei* D4.

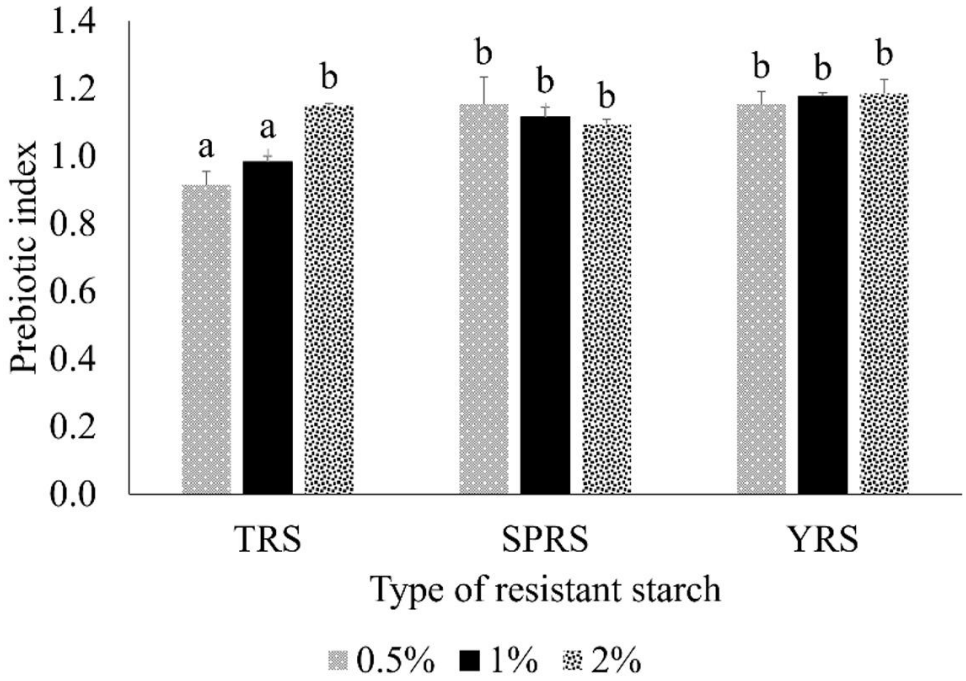

Figure 2. Prebiotic Index of resistant starch from local tubers.

Note: [ab] different lowercase letters indicate significant differences between the types and concentrations of resistant starch, based on Tukey's HSD test ($p < 0.05$).
TRS = taro resistant starch; SPRS = sweet potato resistant starch; YRS = yam resistant starch.

3.3. Probiotic Viability during Gastrointestinal Simulation

The three digestive phases showed a decrease in cell density for *L. paracasei* D4 for all three treatment and control microcapsules (Table 2). In the gastric phase, YRSM showed the lowest decrease in *L. paracasei* D4 viability of 0.96 log CFU/g, from 9.41 ± 0.13 log CFU/g to 8.45 ± 0.06 log CFU/g. Meanwhile, in the gastric phase, the control microcapsule showed the highest decrease in probiotic viability of 2.31 log CFU/g, from 9.39 ± 0.13 log CFU/g to 7.07 ± 0.08 log CFU/g. The better protection of YRSM in maintaining the viability of *L. paracasei* D4 was related to the high amylose content in *D. alata*. A high amylose content produces a more compact and stable crystalline structure of RS (Li, An, et al., 2018). The crystalline structure with a compact double-helix configuration contributes to the physical protection of RS during gastric acid exposure. The use of resistant starch as a microencapsulation matrix has also been reported by Wang et al. (2024), resulting in a lower decrease in probiotic viability, similar to this study. The resistant starch matrix derived from green bananas provided good protection, maintaining the viability of the probiotic cocktail after the simulated gastric test (pH 2), with a 1.49 log CFU/mL decrease in density.

Table 2. Viability of *L. paracasei* D4 in microcapsules throughout the gastrointestinal simulation phases.

Type of microcapsule	Cell Density of L. paracasei D4 (log CFU/g)			
	Pre-GIS	Stomach	Duodenum	Ileum
CM	9.39 ± 0.13^{ns}	7.07 ± 0.08^{a}	6.66 ± 0.39^{a}	6.23 ± 0.10^{a}
TRSM	9.38 ± 0.09	7.74 ± 0.06^{b}	7.33 ± 0.05^{b}	6.87 ±0.07^{b}
SPRSM	9.35 ± 0.13	7.82 ± 0.09^{b}	7.54 ± 0.06^{b}	7.11 ± 0.07^{c}
YRSM	9.41 ±0.13	8.45 ± 0.06^{c}	8.19 ± 0.04^{c}	7.96 ± 0.07^{d}

Note: Data are presented as means ± standard deviations of triplicate measurements in each digestive phase. ^{a,b,c,d} different superscript letters within the same column indicate significant differences among treatments, based on Tukey's HSD test ($p < 0.05$). ns = Mean values within the same column are not significantly different according to Tukey's HSD test ($p > 0.05$). CM = control microcapsule; TRSM = taro resistant starch microcapsule; SPRSM = sweet potato resistant starch microcapsule; YRSM = yam resistant starch microcapsule.

The small intestine simulation phase was divided into two phases: the duodenal and ileal phases. In the small intestine phase, YRSM showed a decrease in the viability of *L. paracasei* D4 of 1.45 log CFU/g, while CM showed a greater decrease in probiotic viability of 3.15 log, from 9.39 ± 0.13 log CFU/g to 6.23 ± 0.10 log CFU/g (Table 2). The addition of SPRS and TRS as encapsulation matrices also showed a positive effect on the microcapsules compared with the microcapsules without RS addition. These results demonstrated that probiotic microencapsulation with RS addition improved cell survival against bile salt exposure and enzymatic activity. In the small intestine phase, RS exhibited slower starch hydrolysis due to its dense crystalline structure, which conferred resistance to digestive enzymes. The stability of RS against digestive enzymatic activity was influenced by a combination of its highly structured molecular architecture and the physical properties of its particles. The compact double-helix crystalline structure effectively blocked access by digestive enzymes to hydrolyze the starch (Shi et al., 2025).

3.4. Microstructural Changes of Microcapsules Before and After Gastrointestinal Simulation

The pre-GIS microcapsules (Figures 3a–d) showed a non-spherical morphology with an irregular and rough surface. It was the sample preparation procedure that caused this surface morphology of the microcapsules, which involved vacuum drying followed by gold coating (Ebel et al., 2024). The surface characteristics of YRSM (Figure 3d) and SPRSM (Figure 3c) seemed almost similar and rougher compared to TRSM. The rougher microcapsule surface was due to the larger granule sizes of yam and sweet potato starches compared to those of taro. The small size of taro starch granules (1–5 μm) (Agama-Acevedo et al., 2011; Nagar, Dash, Rayaguru, Pal, & Nedunchezhiyan, 2021) allowed the granules to fill the small gaps in the alginate matrix more efficiently. Meanwhile, without the addition of resistant starch (CM) (Figure 3a), the microcapsule surface appeared more irregular and rougher. Even though TRSM exhibited a smoother surface, it had a larger pore size (31.72 ± 1.26 μm) than other types of microcapsules (Table 3). In contrast to the findings of this study, a previous study by Zubair et al. (2023) reported that taro starch microcapsules had a smaller pore size than sodium alginate-based microcapsules without added starch.

The pore size range of the pre-GIS microcapsules was 24.31 ± 3.71 µm to 31.72 ± 1.26 µm. This pore size is consistent with previous studies, which reported relatively similar pore sizes. Fructo-oligosaccharides, a prebiotic integrated with alginate-chitosan as the microencapsulation matrix, exhibited a pore size of 17.20 µm (Thinkohkaew et al., 2025).

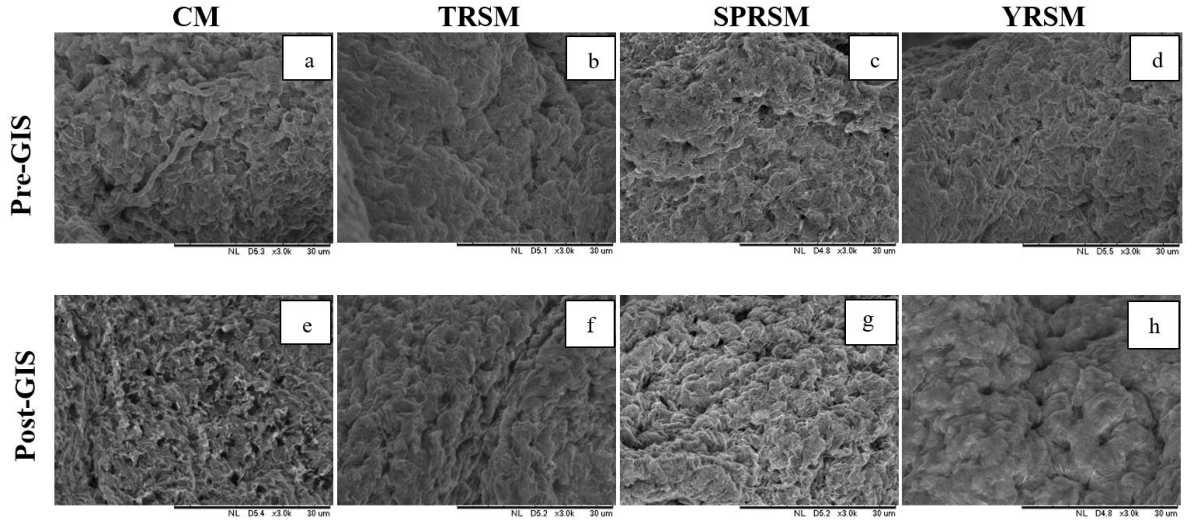

Figure 3. Morphological changes on the surface of *L. paracasei* D4 microcapsules.

Note: Upper panel (pre-GIS): (a) CM, (b) TRSM, (c) SPRSM, (d) YRSM. Lower panel (post-GIS): (e) CM, (f) TRSM, (g) SPRSM, (h) YRSM. CM = control microcapsule; TRSM = taro resistant starch microcapsule; SPRSM = sweet potato resistant starch microcapsule; YRSM = yam resistant starch microcapsule.

SEM images (Figures 3e–h) show changes in the morphology of the microcapsules post-GIS, with increases in surface pore size and number. The surface morphology of CM showed the most significant change, with an increase in the number of pores after the gastrointestinal tract simulation, from six to 74 (Table 3). This indicates that without the addition of RS, the microcapsules were more sensitive to enzymatic activity and pH changes, which could affect the release of probiotics before reaching the colon. Meanwhile, YRSM demonstrated better microcapsule stability than the other microcapsules. There was an increase in pore length in YRSM, from 24.31 ± 3.71 µm to 29.12 ± 15.24 µm, but the increase in the number of pores on the surface was not particularly significant. The changes in the morphology of YRSM post-GIS indicate that this matrix combination maintains microcapsule stability, thereby limiting the formation of new pores on the microcapsule surface.

Table 3. Surface pore changes in microcapsules.

Type of microcapsule	Pore size (µm)		Total pores	
	Pre-GIS	Post-GIS	Pre-GIS	Post-GIS
CM	29.85 ± 4.31^{ab}	31.23 ± 2.19^{ns}	6	74
TRSM	31.77 ± 0.33^{b}	34.73 ± 5.21	4	25
SPRSM	25.06 ± 2.83^{ab}	29.22 ± 2.46	8	14
YRSM	24.11 ± 1.72^{a}	29.12 ± 3.57	10	12

Note: ^{abc} different superscript letters within the same column indicate significant differences among treatments, based on Tukey's HSD test ($p < 0.05$). ^{ns} = Mean values within the same column are not significantly different according to Tukey's HSD test ($p > 0.05$). Pre-GIS = pre-gastrointestinal simulation; post-GIS = post-gastrointestinal simulation; CM = control microcapsule; TRSM = taro resistant starch microcapsule; SPRSM = sweet potato resistant starch microcapsule; YRSM = yam resistant starch microcapsule.

4. CONCLUSION

4.1. Implication

Yam (*D. alata* L.) was found to be more effective than sweet potato and taro as a source of RS3, with the highest amylose (41.92 ± 0.77%) and RS (6.12 ± 0.03%) content after autoclaving–cooling modification. Consistent with its chemical characteristics, YRS also demonstrated good prebiotic activity, yielding the highest PAS (7.58 ± 4.97) and PI (1.19 ± 0.04) values at a 2% concentration, indicating that YRS selectively supports the growth of *L. paracasei* D4. YRSM-based microcapsules provided the best protection for *L. paracasei* D4 during gastrointestinal simulation, with

a lower viability loss (1.45 log CFU/g) compared to control microcapsules (3.15 log CFU/g), as well as the best structural stability, as confirmed by SEM analysis. These findings confirm the potential of yam as a functional ingredient derived from a local Indonesian tuber for the development of commercial synbiotic products.

4.2. Limitation

Probiotic viability testing was conducted on a laboratory scale using *in vitro* simulated gastrointestinal conditions; therefore, the results obtained do not fully represent conditions in the human gastrointestinal tract.

4.3. Future Research Directions

To enhance the application of the RS tuber co-encapsulation matrix as a functional food, additional research is needed to evaluate storage-time analysis and microcapsule viability through *in vivo* gastrointestinal testing.

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Competing Interests: The authors declare that they have no competing interests.
Authors' Contributions: All authors contributed equally to the conception and design of the study. All authors have read and agreed to the published version of the manuscript.
Disclosure of AI use: The authors used Gemini, Claude, and ChatGPT to edit and refine the wording of the manuscript. The authors take full responsibility for the scientific content, data interpretation, and conclusions of this work.
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REFERENCES

Adesokan, M., Alamu, E. O., Otegbayo, B., Asfaw, A., Afolabi, M. O., Fawole, S., . . . Maziya-Dixon, B. (2025). Rapid analysis of starch, sugar, and amylose in fresh yam tubers and boiled yam texture using near-infrared hyperspectral imaging and chemometrics. *Journal of Food Composition and Analysis, 142*, 107425. https://doi.org/10.1016/j.jfca.2025.107425

Afoakwa, E. O., Polycarp, D., Budu, A. S., Mensah-Brown, H., & Otoo, E. (2013). Variability in biochemical composition and cell wall constituents among seven varieties in Ghanaian yam (Dioscorea sp.) germplasm. *African Journal of Food, Agriculture, Nutrition and Development, 13*(4), 8106–8127. https://doi.org/10.18697/ajfand.59.13280

Agama-Acevedo, E., Garcia-Suarez, F. J., Gutierrez-Meraz, F., Sanchez-Rivera, M. M., San Martin, E., & Bello-Pérez, L. A. (2011). Isolation and partial characterization of Mexican taro (Colocasia esculenta L.) starch. *Starch-Stärke, 63*(3), 139-146. https://doi.org/10.1002/star.201000113

Andrade, L. A., Barbosa, N. A., & Pereira, J. (2017). Extraction and properties of starches from the non-traditional vegetables yam and taro. *Polímeros, 27*(2), 151–157. https://doi.org/10.1590/0104-1428.04216

AOAC International. (2005). *Official methods of analysis of AOAC International* (18th ed.). Gaithersburg, MD: AOAC International.

Aprianita, A., Vasiljevic, T., Bannikova, A., & Kasapis, S. (2014). Physicochemical properties of flours and starches derived from traditional Indonesian tubers and roots. *Journal of Food Science and Technology, 51*(12), 3669-3679. https://doi.org/10.1007/s13197-012-0915-5

Boahemaa, L. V., Dzandu, B., Amissah, J. G. N., Akonor, P. T., & Saalia, F. K. (2024). Physico-chemical and functional characterization of flour and starch of taro (Colocasia esculenta) for food applications. *Food and Humanity, 2*, 100245. https://doi.org/10.1016/j.foohum.2024.100245

Bojarczuk, A., Skąpska, S., Khaneghah, A. M., & Marszałek, K. (2022). Health benefits of resistant starch: A review of the literature. *Journal of Functional Foods, 93*, 105094. https://doi.org/10.1016/j.jff.2022.105094

Buenaventura, A. A., Reginio, J. F. C., Duque, S. M. M., Agustin, R. C. L., Laviña, W. A., & Mopera, L. E. (2025). The functional dessert: Purple yam jam starch digestibility and prebiotic potential. *Applied Food Research*, *5*(2), 101464. https://doi.org/10.1016/j.afres.2025.101464

Chiranthika, N. N. G., Chandrasekara, A., & Gunathilake, K. D. P. P. (2022). Physicochemical characterization of flours and starches derived from selected underutilized roots and tuber crops grown in Sri Lanka. *Food Hydrocolloids*, *124*, 107272. https://doi.org/10.1016/j.foodhyd.2021.107272

Cornejo-Ramírez, Y. I., Martínez-Cruz, O., Del Toro-Sánchez, C. L., Wong-Corral, F. J., Borboa-Flores, J., & Cinco-Moroyoqui, F. J. (2018). The structural characteristics of starches and their functional properties. *CyTA - Journal of Food*, *16*(1), 1003-1017. https://doi.org/10.1080/19476337.2018.1518343

Cui, Q., Mahfouzi, M., Zhang, H., Gao, R., & Guo, Z. (2024). Characteristics of sweet potato starch particles modified by various methods for use in food-grade Pickering emulsion. *LWT*, *210*, 116831. https://doi.org/10.1016/j.lwt.2024.116831

da Silva, M. N., Tagliapietra, B. L., do Amaral Flores, V., & dos Santos Richards, N. S. P. (2021). In vitro test to evaluate survival in the gastrointestinal tract of commercial probiotics. *Current Research in Food Science*, *4*, 320-325. https://doi.org/10.1016/j.crfs.2021.04.006

de Albuquerque, T. M. R., Borges, C. W. P., Cavalcanti, M. T., dos Santos Lima, M., Magnani, M., & de Souza, E. L. (2020). Potential prebiotic properties of flours from different varieties of sweet potato (Ipomoea batatas L.) roots cultivated in Northeastern Brazil. *Food Bioscience*, *36*, 100614. https://doi.org/10.1016/j.fbio.2020.100614

Ebel, F., Ramírez-Reveco, A., Strobel, P., Wagenknecht, L., Rodríguez, N., Bosch, P., & Rivarola, C. (2024). Optimized protocol for high-vacuum scanning electron microscopy analysis of polyacrylamide hydrogel-attached sperm cells in a binary system. *Microscopy Research and Technique*, *87*(5), 1122-1127. https://doi.org/10.1002/jemt.24502

Englyst, H. N., Kingman, S. M., & Cummings, J. H. (1992). Classification and measurement of nutritionally important starch fractions. *European Journal of Clinical Nutrition*, *46*(Suppl. 2), S33–S50.

Erickson, J. M., Carlson, J. L., Stewart, M. L., & Slavin, J. L. (2018). Fermentability of novel type-4 resistant starches in in vitro system. *Foods*, *7*(2), 18. https://doi.org/10.3390/foods7020018

Etchepare, M. d. A., Raddatz, G. C., Flores, d. M. É. M., Zepka, L. Q., Jacob-Lopes, E., Barin, J. S., ... de Menezes, C. R. (2016). Effect of resistant starch and chitosan on survival of Lactobacillus acidophilus microencapsulated with sodium alginate. *LWT*, *65*, 511-517. https://doi.org/10.1016/j.lwt.2015.08.039

Etchepare, M. d. A., Barin, J. S., Cichoski, A. J., Jacob-Lopes, E., Wagner, R., Fries, L. L. M., & Menezes, C. R. d. (2015). Microencapsulation of probiotics using sodium alginate. *Ciência Rural*, *45*(7), 1319-1326. https://doi.org/10.1590/0103-8478cr20140938

Feng, H., Cheng, B., Lim, J., Li, B., Li, C., & Zhang, X. (2024). Advancements in enhancing resistant starch type 3 (RS3) content in starchy food and its impact on gut microbiota: A review. *Comprehensive Reviews in Food Science and Food Safety*, *23*(3), e13355. https://doi.org/10.1111/1541-4337.13355

Figueroa-González, I., Rodríguez-Serrano, G., Gómez-Ruiz, L., García-Garibay, M., & Cruz-Guerrero, A. (2019). Prebiotic effect of commercial saccharides on probiotic bacteria isolated from commercial products. *Food Science and Technology*, *39*(3), 747-753. https://doi.org/10.1590/fst.07318

Goderska, K., & Kozłowski, P. (2021). Evaluation of microencapsulated synbiotic preparations containing lactobionic acid. *Applied Biochemistry and Biotechnology*, *193*(11), 3483-3495. https://doi.org/10.1007/s12010-021-03622-9

Goh, C. H., Heng, P. W. S., & Chan, L. W. (2012). Alginates as a useful natural polymer for microencapsulation and therapeutic applications. *Carbohydrate Polymers*, *88*(1), 1-12. https://doi.org/10.1016/j.carbpol.2011.11.012

Hapsari, L., Mas'udah, S., & Fauziah, F. (2023). Traditional knowledge in conservation and farming practices of water yam (Dioscorea alata L.): Lessons learned from East Javanese community. *International Journal of Conservation Science*, *14*(4), 1573-1590. https://doi.org/10.36868/IJCS.2023.04.21

Hill, C., Guarner, F., Reid, G., Gibson, G. R., Merenstein, D. J., Pot, B., . . . Sanders, M. E. (2014). The International Scientific Association for Probiotics and Prebiotics consensus statement on the scope and appropriate use of the term probiotic. *Nature Reviews Gastroenterology & Hepatology*, *11*(8), 506-514. https://doi.org/10.1038/nrgastro.2014.66

Horison, R., & Surini, S. (2024). Harnessing the potential of several local Indonesian tuber-derived starches as pharmaceutical excipients. *Pharmaceutical Sciences and Research*, *11*(2), 68–87. https://doi.org/10.7454/psr.v11i2.1385

Huebner, J., Wehling, R. L., & Hutkins, R. W. (2007). Functional activity of commercial prebiotics. *International Dairy Journal*, *17*(7), 770-775. https://doi.org/10.1016/j.idairyj.2006.10.006

Jagtiani, E., & Adsare, S. (2022). Microencapulsation: Probiotics, prebiotics, and nutraceuticals. *Journal of Nanotechnology and Nanomaterials*, *3*(1), 34-60. https://doi.org/10.33696/Nanotechnol.3.030

Jan, T., Negi, R., Hilal, A., Kaur, S., Kour, D., Khan, S. S., . . . Ahmed, N. (2025). Microencapsulation techniques for probiotic formulations: Current scenario and future perspective. *Journal of Food Quality*, *2025*(1), 6738124. https://doi.org/10.1155/jfq/6738124

Jatmiko, Y. D., Suharjono, S., Ardyati, T., Mustafa, I., Mustamin, A., Puja, L. R., . . . Atho'illah, M. F. (2025). Unlocking the probiotic with antioxidant-rich potential from wine coffee: In vitro screening and characterization. *Coffee Science*, *20*, e202341. https://doi.org/10.25186/.v20i.2341

Jayakody, L., Hoover, R., Liu, Q., & Donner, E. (2007). Studies on tuber starches. II. Molecular structure, composition and physicochemical properties of yam (Dioscorea sp.) starches grown in Sri Lanka. *Carbohydrate Polymers*, *69*(1), 148-163. https://doi.org/10.1016/j.carbpol.2006.09.024

Kim, H.-J., Woo, K. S., Lee, H.-U., Nam, S. S., Lee, B. W., Kim, M. Y., . . . Lee, B. (2020). Physicochemical characteristics of starch in sweet potato cultivars grown in Korea. *Preventive Nutrition and Food Science*, *25*(2), 212-218.

Kustyawati, M. E., Nurlita, E. M., Fadhallah, E. G., & Rizal, S. (2022). Prebiotic activity of Lactobacillus casei grown on medium containing of Hylocereus undatus extract and its use in the fermentation of goat's milk kefir. *Biodiversitas Journal of Biological Diversity*, *23*(12), 6513-6519. https://doi.org/10.13057/biodiv/d231249

Latif, A., Shehzad, A., Niazi, S., Zahid, A., Ashraf, W., Iqbal, M. W., . . . Korma, S. A. (2023). Probiotics: Mechanism of action, health benefits and their application in food industries. *Frontiers in Microbiology*, *14*, 1216674. https://doi.org/10.3389/fmicb.2023.1216674

Lee, Y. R., Lee, H.-B., Kim, Y., Shin, K.-S., & Park, H.-Y. (2023). Prebiotic and anti-adipogenic effects of radish green polysaccharide. *Microorganisms*, *11*(7), 1862. https://doi.org/10.3390/microorganisms11071862

Li, M., & Chou, C. F. (2018). Effect of different yam varieties and addition on resistant starch type III formation and physical properties of steamed rice bowl cake. *Journal of Food and Nutrition Research*, *6*(11), 682–688. https://doi.org/10.12691/jfnr-6-11-1

Li, T., An, F., Teng, H., Huang, Q., Zeng, F., & Song, H. (2018). Comparison of structural features and in vitro digestibility of purple yam (Dioscorea alata L.) resistant starches by autoclaving and multi-enzyme hydrolysis. *Food Science and Biotechnology*, *27*(1), 27-36. https://doi.org/10.1007/s10068-017-0206-z

Li, T., Chen, L., Xiao, J., An, F., Wan, C., & Song, H. (2018). Prebiotic effects of resistant starch from purple yam (Dioscorea alata L.) on the tolerance and proliferation ability of Bifidobacterium adolescentis in vitro. *Food & Function*, *9*(4), 2416-2425. https://doi.org/10.1039/c7fo01919j

Lopulalan, C. G. C., Marseno, D. W., Marsono, Y., & Pranoto, Y. (2023). Chemical characterization, total starch, amylose and resistance starch of local cocoyam in Maluku, Indonesia. *Food Research*, *7*(1), 230-235.

Luca, L., & Oroian, M. (2021). Influence of different prebiotics on viability of Lactobacillus casei, Lactobacillus plantarum and Lactobacillus rhamnosus encapsulated in alginate microcapsules. *Foods*, *10*(4), 710. https://doi.org/10.3390/foods10040710

Lyu, R., Ahmed, S., Fan, W., Yang, J., Wu, X., Zhou, W., . . . Wang, H. (2021). Engineering properties of sweet potato starch for industrial applications by biotechnological techniques including genome editing. *International Journal of Molecular Sciences*, *22*(17), 9533. https://doi.org/10.3390/ijms22179533

Maharani, R. S., & Arzani, L. D. P. (2024). Evaluation of sensory attributes of soft candy using taro leaf extract as influenced by variations of gelatin and glucose syrup. *Jurnal Teknologi dan Mutu Pangan*, *3*(1), 22-30. https://doi.org/10.30812/jtmp.v3i1.4177

Maretta, D., Sobir, Helianti, I., Purwono, & Santosa, E. (2021). Current status of taro (Colocasia esculenta) utilization as local food diversification toward climate resilience in Indonesia. *IOP Conference Series: Earth and Environmental Science*, *913*(1), 012027. https://doi.org/10.1088/1755-1315/913/1/012027

Moghanjougi, M. Z., Bari, R. M., Khaledabad, A. M., Amiri, S., & Almasi, H. (2021). Microencapsulation of Lactobacillus acidophilus LA-5 and Bifidobacterium animalis BB-12 in pectin and sodium alginate: A comparative study on viability, stability, and structure. *Food Science & Nutrition*, *9*(9), 5103-5111. https://doi.org/10.1002/fsn3.2470

Muhammad, Z., Ramzan, R., Zhang, R., & Zhang, M. (2021). Resistant starch-based edible coating composites for spray-dried microencapsulation of Lactobacillus acidophilus, comparative assessment of thermal protection, in vitro digestion and physicochemical characteristics. *Coatings*, *11*(5), 587. https://doi.org/10.3390/coatings11050587

Nagar, C. K., Dash, S. K., Rayaguru, K., Pal, U. S., & Nedunchezhiyan, M. (2021). Isolation, characterization, modification and uses of taro starch: A review. *International Journal of Biological Macromolecules*, *192*, 574-589. https://doi.org/10.1016/j.ijbiomac.2021.10.041

Ndisya, J. M., Mbuge, D. O., Gitau, A. N., & Sturm, B. (2026). Advances in preservation and transformation of starchy African tubers: Current status, challenges, and opportunities. In T. S. Workneh, D. O. Mbuge, & G. Bultosa (Eds.), *Processing, nutrition and value addition principles of neglected and underutilized African foods* (pp. 127–145). Amsterdam, the Netherlands: Elsevier. https://doi.org/10.1016/B978-0-443-33433-7.00012-5

Nguon, O., Lagugné-Labarthet, F., Brandys, F. A., Li, J., & Gillies, E. R. (2018). Microencapsulation by in situ polymerization of amino resins. *Polymer Reviews*, *58*(2), 326-375. https://doi.org/10.1080/15583724.2017.1364765

Ochoa, S., & Osorio-Tobón, J. F. (2024). Isolation and characterization of starch from the purple yam (Dioscorea alata) anthocyanin extraction residue obtained by ultrasound-assisted extraction. *Waste and Biomass Valorization*, *15*(1), 379-389. https://doi.org/10.1007/s12649-023-02155-y

Palacio, M. I., Etcheverría, A. I., & Manrique, G. D. (2014). Fermentation by Lactobacillus paracasei of galactooligosaccharides and low-molecular-weight carbohydrates extracted from squash (Curcubita maxima) and lupin (Lupinus albus) seeds. *Journal of Microbiology, Biotechnology and Food Sciences*, *3*(4), 329-332.

Rana, A., Taneja, N. K., Raposo, A., Alarifi, S. N., Teixeira-Lemos, E., Lima, M. J., . . . Dhewa, T. (2024). Exploring prebiotic properties and its probiotic potential of new formulations of soy milk-derived beverages. *Frontiers in Microbiology*, *15*, 1404907. https://doi.org/10.3389/fmicb.2024.1404907

Rattanakiat, S., Pulbutr, P., Khunawattanakul, W., Sungthong, B., & Saramunee, K. (2020). Prebiotic activity of polysaccharides extracted from Jerusalem artichoke tuber and development of prebiotic granules. *Pharmacognosy Journal*, *12*(6), 1402-1411. https://doi.org/10.5530/pj.2020.12.194

Rifa'i, M., Atho'illah, M. F., Arifah, S. N., Suharto, A. R., Fadhilla, A. N., Sa'adah, N. A. M., … Jatmiko, Y. D. (2025). Physicochemical and functional optimization of probiotic yogurt with encapsulated Lacticaseibacillus paracasei E1 enriched with green tea using Box–Behnken design. *Applied Food Research*, *5*(1), 100690. https://doi.org/10.1016/j.afres.2024.100690

Rosida, Harijono, Teti, E., & Endang, S. (2016). Hypoglycemic effect of modified water yam flour (Dioscorea alata) on diabetic Wistar rats (Rattus norvegicus). *Journal of Food and Nutrition Research*, *4*(1), 20–25.

Sarvari, F., Mortazavian, A. M., & Fazeli, M. R. (2014). Biochemical characteristics and viability of probiotic and yogurt bacteria in yogurt during the fermentation and refrigerated storage. *Applied Food Biotechnology*, *1*(1), 55-61. https://doi.org/10.22037/afb.v1i1.7125

Sbehat, M., Mauriello, G., & Altamimi, M. (2022). Microencapsulation of probiotics for food functionalization: An update on literature reviews. *Microorganisms*, *10*(10), 1948. https://doi.org/10.3390/microorganisms10101948

Setiarto, R. H. B., Kusumaningrum, H. D., Jenie, B. S. L., Khusniati, T., & Sulistiani, S. (2020). Modified taro starch as alternative encapsulant for microencapsulation of Lactobacillus plantarum SU-LS 36. *Czech Journal of Food Sciences, 38*(5), 293–300. https://doi.org/10.17221/2/2020-CJFS

Setiarto, R. H. B., Kusumaningrum, H. D., Jenie, B. S. L., Khusniati, T., Widhyastuti, N., & Sulistiani. (2019). *Effect of heat process for prebiotic properties of taro starch (Colocasia Esculenta L. Schott).* Paper presented at the Proceeding 2nd SEAFAST Int. Seminar (2nd SIS 2019): Facing Future Challenges: Sustainable Food Safety, Quality and Nutrition (pp. 77–83), Bogor, Indonesia: SciTePress.

Shi, J., Zeng, K., Yang, Y., An, X., Guo, J., Hu, Y., . . . Liu, S. (2025). Structural changes of type 3 resistant starch during in vivo digestion using a duodenum and ileal cannulated miniature pig model. *Carbohydrate Polymers, 360*, 123608. https://doi.org/10.1016/j.carbpol.2025.123608

Silva, G. M. d. S., Veloso, C. M., Santos, L. S., Melo Neto, B. A. d., Fontan, R. d. C. I., & Bonomo, R. C. F. (2020). Extraction and characterization of native starch obtained from the inhambu tuber. *Journal of Food Science and Technology, 57*(5), 1830-1839. https://doi.org/10.1007/s13197-019-04216-4

Silva, P. B., Garcia, S., Baldo, C., & Celligoi, M. A. P. C. (2017). Prebiotic activity of fructooligosaccharides produced by Bacillus subtilis natto CCT 7712. *Acta Alimentaria, 46*(2), 145-151. https://doi.org/10.1556/066.2016.0004

Simsek, S., & El, S. N. (2012). Production of resistant starch from taro (Colocasia esculenta L. Schott) corm and determination of its effects on health by in vitro methods. *Carbohydrate Polymers, 90*(3), 1204-1209. https://doi.org/10.1016/j.carbpol.2012.06.039

Soral-Śmietana, M., Wronkowska, M., Biedrzycka, E., Bielecka, M., & Ocicka, K. (2005). Native and physically-modified starches–utilization of resistant starch by bifidobacteria (in vitro). *Polish Journal of Food and Nutrition Sciences, 55*(3), 273-279.

Sultana, K., Godward, G., Reynolds, N., Arumugaswamy, R., Peiris, P., & Kailasapathy, K. (2000). Encapsulation of probiotic bacteria with alginate–starch and evaluation of survival in simulated gastrointestinal conditions and in yoghurt. *International Journal of Food Microbiology, 62*(1-2), 47-55. https://doi.org/10.1016/S0168-1605(00)00380-9

Tanimola, A. R., Otegbayo, B. O., & Akinoso, R. (2022). Physicochemical properties of yam starches from fifty-five lines of Dioscorea species. *Food Research, 6*(3), 49-61.

Thinkohkaew, K., Jonjaroen, V., Niamsiri, N., McClements, D. J., Panya, A., Suppavorasatit, I., & Potiyaraj, P. (2025). Fabrication of synbiotic carbohydrate polymer-based microcapsules: Effect of prebiotics on probiotic viability during freeze-drying, gastrointestinal transit and storage. *Carbohydrate Polymers, 359*, 123582. https://doi.org/10.1016/j.carbpol.2025.123582

Thuy, N. M., Too, B. C., Vuong, K. M., Lan, P. T. T., Tuyen, P. T. T., Tram, N. B., … Tai, N. V. (2022). Resistant starch in various starchy vegetables and the relationship with its physical and chemical characteristics. *Journal of Applied Biology & Biotechnology, 10*(1), 181-188. https://doi.org/10.7324/JABB.2021.100122

Torp, A. M., Bahl, M. I., Boisen, A., & Licht, T. R. (2022). Optimizing oral delivery of next generation probiotics. *Trends in Food Science & Technology, 119*, 101-109. https://doi.org/10.1016/j.tifs.2021.11.034

Walsen, A., Polnaya, F., Lesilolo, M. K., Rehatta, H., & Lawalata, I. J. (2020). The appropriate of plant propagation technology of yam (Dioscorea alata L) to reduce multiplication ratio. *Journal of Physics: Conference Series, 1463*(1), 012029. https://doi.org/10.1088/1742-6596/1463/1/012029

Wang, J.-L., Yeh, C.-H., Huang, S.-H., Wu, L. S.-H., & Chen, M. C.-M. (2024). Effects of resistant-starch-encapsulated probiotic cocktail on intestines damaged by 5-fluorouracil. *Biomedicines, 12*(8), 1912. https://doi.org/10.3390/biomedicines12081912

Zang, Z., Gong, X., Cao, L., Ni, H., & Chang, H. (2024). Resistant starch from yam: Preparation, nutrition, properties and applications in the food sector. *International Journal of Biological Macromolecules, 273*, 133087. https://doi.org/10.1016/j.ijbiomac.2024.133087

Zanjani, M. A. K., Tarzi, B. G., Sharifan, A., & Mohammadi, N. (2014). Microencapsulation of probiotics by calcium alginate-gelatinized starch with chitosan coating and evaluation of survival in simulated human gastro-intestinal condition. *Iranian Journal of Pharmaceutical Research, 13*(3), 843–852. https://doi.org/10.22037/ijpr.2014.1550

Zhao, X., Andersson, M., & Andersson, R. (2018). Resistant starch and other dietary fiber components in tubers from a high-amylose potato. *Food Chemistry*, *251*, 58-63. https://doi.org/10.1016/j.foodchem.2018.01.028

Zheng, G., & Mao, J. (2026). Mechanistic two-pathway modeling of substrate inhibition in lactic acid bacteria for enhanced fermentation control. *Quantitative Biology*, *14*(1), e70019. https://doi.org/10.1002/qub2.70019

Zheng, H., Shi, F., Li, J., Bian, X., Wu, S., & Li, X. (2025). The dose-dependent influence of type 2 resistant starch on gut microbial communities and metabolic outputs: An in vitro simulation. *Foods*, *14*(18), 3255. https://doi.org/10.3390/foods14183255

Zhu, F. (2015). Isolation, composition, structure, properties, modifications, and uses of yam starch. *Comprehensive Reviews in Food Science and Food Safety*, *14*(4), 357-386. https://doi.org/10.1111/1541-4337.12134

Zubair, W., Afzaal, M., Saeed, F., Ahmed, A., Ateeq, H., Akram, N., . . . Teferi Asres, D. (2023). Survivability of Lactobacillus rhamnosus under stressed conditions as affected by taro starch (Colocasia esculenta) encapsulation. *International Journal of Food Properties*, *26*(2), 3465-3476. https://doi.org/10.1080/10942912.2023.2286892