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The Physical, Chemical, Microbiological, Antibacterial and Prebiotic Characteristics of Fermented Porang Flour with Addition of Bacteria and Yeast

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Abstract. Porang (*Amorphophallus oncophylus*) tubers are one of the export commodities due to their glucomannan content. This study aimed to investigate the characteristics of water fermentation and the quality of porang fermented flour with the addition of *Lactobacillus plantarum* (LP), *Bacillus subtilis* (BS), and *Saccharomyces cerevisiae* (SC). Fermentation was conducted using four treatments, namely LP+BS, LP+SC, BS+SC which consisted of 10%v/v of inoculum, respectively, and LP+BS+SC which consisted of 7%v/v LP, 7% BSv/v, and 6%SCv/v. During fermentation, pH, total acidity, and total microorganisms were observed. Proximate, glucomannan content, starch content, fiber content, and antibacterial and prebiotics characteristics were observed for fermented porang flour. The results showed that pH decreased while total acidity increased during fermentation. Total aerobic bacteria had the highest population at 24 hours of fermentation while lactic acid bacteria had the highest population at 24- and 48-hour fermentation. Yeast reached the highest population at 72 hours of fermentation. Porang fermented flour with the addition of BS+SC contained the highest glucomannan content, carbohydrate, and fiber content. Moreover, the flour with BS+SC had an antibacterial effect against *Escherichia coli* and *Staphylococcus aureus* and supported the growth of *Lactobacillus acidophilus*. Porang fermented flour which is processed with the addition of a mixed culture of BS and SC can be developed to improve the nutritional and functional properties of porang flour.

Keywords: bacteria; fermentation; flour; porang; yeast

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INTRODUCTION

One of the processed products of porang tubers is porang flour. Processing porang to be flour is done to increase the economic value of porang and prevent damage to porang tubers. The quality of porang flour is determined by the high glucomannan content (Budiastra & Noviyanti, 2023). Glucomannan has been used in various fields such as food, health, cosmetics, and flavor binders in seasonings (Behera & Ray, 2016; Devaraj et al. 2019; Nurlela et al. 2019; Widjanarko et al. 2022; Budiastra & Noviyanti 2023). Glucomannan content in porang tubers has some characteristics such as high fiber content, low calories, hydrocolloid, and low cholesterol (Widjanarko et al. 2022; Suryana et al. 2022). Due to these reasons, porang is suitable for use as a substitute for staple foods such as rice (Wahidah et al., 2021). Moreover, glucomannan is known as generally recommended as safe (GRAS) so it is safe to consume (Behera and Ray 2016; Gómez et al. 2017). Some previous studies showed that glucomannan also had prebiotic capability (Wardhani et al., 2017; Anggela et al., 2022).

Glucomannan is found in parenchyma tissue and wrapped in starch in porang or konjac tubers (Nurlela et al. 2021; Widjanarko et al. 2022). Starch is an impurity compound in the extraction of glucomannan (Wardhani et al. 2019; Nurlela et al. 2021; Widjanarko et al. 2022). Microorganisms have some enzymes to degrade starch such as amylase. Several microorganisms have amylase activity and have been used as starters in the food industry, namely *Bacillus subtilis*, *Lactobacillus plantarum,* and *Saccharomyces cerevisiae*.

Bacillus subtilis is a kind of bacteria that has amylase (David et al., 2018; Hu & Liu, 2021). *B. subtilis* does not produce toxins, is easy to grow, does not need expensive substrates, and does not produce any metabolic side product (Andriani et al., 2017). *B.subtilis* has been studied to be developed into a probiotic, such as a probiotic for shrimp (Andriani et al., 2017). *B.subtilis* also excretes antimicrobial compounds and has the potential to be used as a food preservative (Caulier et al., 2019). *B.subtilis* has been used as a starter for the fermentation of natto (Afzaal et al., 2022). *L.plantarum* has amylase activity (Hattingh et al., 2015). *L.plantarum* produces bacteriocins that have antimicrobial effects (Kumar et al., 2016). *L.plantarum* has been used as a starter for some fermented foods, namely fermented taro tubers (Wewo et al., 2018), sauerkraut (Zubaidah et al., 2020), and sour cassava (Penido et al., 2018). *L.plantarum* is also known as a probiotic (Fidanza et al., 2021). *Saccharomyces cerevisiae* has amylase activity. *S.cerevisiae* has been used as a starter in the beer fermentation and bread industry (Parapouli et al., 2020). *S.cerevisiae* has antimicrobial and potential probiotics (Fakruddin et al., 2017; Makky et al., 2021).

Fermentation is an ancient technique in processing food that uses the capability of microorganisms to degrade raw materials into be desired product (Feng et al., 2018; Mani, 2018; Sharma et al., 2020). Fermentation techniques have been applied to some fermented flour, namely sour cassava (Penido et al., 2018), cassava-modified flour (Apriliani & Mulyadi, 2022), wheat fermented flour or sourdough (Zhao et al., 2016; Dimidi et al., 2019). Some characteristics of foods change due to the activity of microorganisms involved in fermentation. Fermented foods have some benefits such as preserving foods and increasing the nutritive, namely amino acids, vitamins, and minerals (Dimidi et al., 2019; Sharma et al., 2020). These compounds support the growth of probiotics and stimulate the prebiotic functions, consequently improving the functional properties of food (Cuamatzin-garcía et al., 2022). Some fermented foods contain probiotics such as kimchi (Song et al., 2023) and hentak (Singh et al., 2018). A prebiotic is defined as a non-digestible and non-hydrolyzable carbohydrate that encourages the growth of good bacteria in the intestinal tract (Wardhani et al., 2017)

The fermented porang flour with the addition of *Lactobacillus bulgaricus* contained lower starch than without fermentation (Nur'aini et al., 2021). The treatments of mixing cultures of *L. plantarum*, *B. subtilis,* and *S.cerevisiae* in processing porang tubers had not been investigated. This research aimed to investigate the effect of adding a mixture starter of *L.plantarum*, *B. subtilis,* and *S.cerevisiae* on the characteristics physical, chemical, and microbiological of fermentation water during fermentation. Moreover, this study aimed to investigate the nutrition, antibacterial, and prebiotic characteristics of fermented porang flour after 72 hours of fermentation

METHODS

Preparation of starter

Bacillus subtilis (BS) was cultivated in Nutrient Broth (NB) media, *Lactobacillus plantarum* (LP) was cultivated in MRS broth, *Saccharomyces cerevisiae* (SC) was cultivated in Potato Dextrose Broth (PDB) at pH 4. A total of 2 loops of inoculation needles of 24 hours old pure cultures of *B.subtilis* in slant agar were taken and dipped in 100 mL of sterilized Nutrient Broth liquid media. For *L. plantarum* and *S.cerevisiae*, 2 loops of inoculation needle of 48-hour-old pure cultures of *L.plantarum* and *S.cerevisiae* in slant agar were taken and dipped in 100 mL of sterilized MRS liquid media for *L.plantarum* and PDB for *S.cerevisiae*. *B.subtilis* was incubated at room temperature for 24 hours while *L.plantarum* and *S.cervisiae* were incubated for 48 hours. After 24 hours incubation for *B.subtilis* and 48 hours for *L.plantarum* and *S.cerevisiae*, the growth of bacteria and yeast in the medium were centrifuged at 10,000 rpm for 10 minutes. The supernatants were discarded. The cells were washed using distilled water and centrifuged again at 10,000 rpm for 10 minutes. Then the cells were added to distilled water until McFarland 0.5 turbidity was obtained.

Fermentation of porang

Peeled, sliced, and washed porang tubers were weighed 1 kg and soaked with 1.5 L sterile distilled water in a closed plastic container. Stater of bacteria and yeast for all treatments were added to the container. Fermentation was conducted using four treatments, namely LP+BS, LP+SC, BS+SC which consisted of 10%v/v of inoculum, respectively, and LP+BS+SC which consisted of 7%v/v LP, 7% BSv/v, and 6%SCv/v. Each treatment was done in three replicates. Fermentation was carried out for 72 hours. Every 24 hours, 150 mL of water fermentation was taken

for further analysis. After 72 hours, fermented porang tubers were drained and dried using a food dehydrator at 50°C for 24 hours. Dried fermented porang tubers were ground and sieved using 90 mesh sieves. The flour yielded was mentioned as fermented porang flour.

pH and total acidity

pH was measured using a pH meter (Ohaus ST 2100-F, USA). Total acidity was determined using titration of 0.1 N NaOH (Kaminarides et al., 2019).

Total microorganism

Bacterial and yeast enumeration was done according to the total plate count method. 100 µL of porang fermentation water from serial dilution tubes from 10^{-1} until 10^{-6} dilution were spread on to Nutrient Agar (NA), de Man Ragose Agar (MRS agar), and Potato Dextrose Agar (PDA) for cultivated aerobic bacteria, lactic acid bacteria and yeast, respectively. NA and MRS agar were incubated at 37°C for 24 hours and 48 hours for aerobic bacteria and lactic acid bacteria, respectively. PDA was incubated at 25°C for 72 hours for cultivation of yeast. The total colonies growing on the agar plates were counted.

Proximate analysis

Moisture was determined by the thermogravimetry method (oven drying methods) (AOAC, 2006). Ash content was measured by thermogravimetry method (furnace method) (AOAC, 2006). The crude protein was measured by the Kjeldahl method principle with a 6.25 conversion factor using the Foss Tecator Kjeltec 8400 instrument (Persson et al., 2008). Crude fat was determined by the hydrolysis/Weibull method (Alika & Atma 2018). Carbohydrate content was calculated by subtracting 100% from the sum of water, protein, ash, and fat contents. All of the aforementioned measurements were performed in triplicates

Glucomannan content

Glucomannan content was measured using DNS according to Chua et al., (2012) . Briefly, there were some steps to measure the glucomannan quantitatively, namely glucomannan extraction, construction of glucose curve standard, extraction of glucomannan extract, extraction of glucomannan hydrolysate, and absorbance measurement.

Strach

Starch content was determined by enzymatic colorimetric method (Zhu et al. 2016). 100 mg of flour was added to a test tube. Next, 0.2 mL of ethanol solution (80%, v/v) was added into the tube. Next, 3.0 mL of thermostable α-amylase (Megazyme, USA) was immediately added, and the tubes were boiled for 6 min and stirred at 2-min intervals. Tubes were then placed in a 50°C water bath for 5 min. Next, 0.1 mL of amyloglucosidase (Megazyme, USA) was added to each tube. Tubes were then stirred and incubated for 30 min. 10 mL of distilled water was poured into each tube and followed by centrifugation at 1,800 × g for 10 min at room temperature. Next, 1.0 mL of aliquots from the supernatant was diluted to 10 mL with distilled water. Then, 0.1 mL of this diluted solution was placed into a clean test tube. A total of 3 mL of glucose oxidase reagent (Diagnostic System International, Germany) was added to each tube and incubated at 50°C for 20 min. For blanks, 0.1 mL of water was used instead of 0.1 mL of diluted solution, and the other reagents were added with the same quantity. Samples were read for absorbance at 510 nm.

Crude fiber

The crude fiber was determined by gravimetry methods (Dwiloka et al. 2019). 1 gram (x) of samples was mixed with 50 ml of H_2SO_4 0.3 N and extracted for 30 min. Then, the extraction result was added with 25 ml of NaOH 1.5 N and extracted again for 30 min. Whatman filter paper number 41 was heated at 105°C for 1 hour and then weighed (a). The extracted extract was then filtered with heated Whatman 41 paper. The filtered sample is then washed successively with 50 ml of hot water, 50 ml of H_2SO_4 0.3 N, 50 ml of hot water, and 25 ml of 70% alcohol. Filter paper and the contents of the sample were put in a porcelain dish and then dried in an oven at 105° C for 6 hours. The sample was cooled in the desiccator for 15 min then weighed (Y). After weighing, the sample was put in a furnace temperature of 550°C for 6 hours then cooled in a desiccator, and weighed (Z).

Calculation of crude fiber by the formula:

$$\%\, Crude\ Fiber = \frac{Y - Z - a}{x} \times 100$$

Antibacterial activity

Antibacterial activity has been carried out according to Helmi et al., (2015). The antibacterial

activity of porang fermented flour was determined by the disc diffusion method. Porang fermented flour was dissolved in aquadest until getting the concentration (1%, 10%, and 50%). A total of 100 µL bacteria suspension with turbidity Mc. Farland 0.5 was spread onto Petri dishes (90 mm on each side). Then, a 6 mm diameter of the blank disc that had been soaked in porang fermented flour with different concentrations for 5 minutes was placed on the surface of the medium. Antibiotic kanamycin as positive control was prepared in the same treatment with porang fermented flour. Petri dishes were incubated at 37°C for 24 hours. The clear zone surrounding the disc represented the antibacterial activity. The diameter of the clear zone was measured using a digital Vernier Caliper. The clear zone was calculated by subtracting the total inhibition by the diameter of the paper disc (6 mm).

Prebiotic activity

Prebiotic activity was observed by total viable count according to Wardhani et al., (2017) with modification. Fermented porang flour was used as a substrate to support the growth of *Lactobacillus acidophilus* as a probiotic. Fermented porang flour (10 grams), MRS Broth media (90mL), and *L.acidophilus* were placed in an Erlenmeyer tube and incubated at 37°C for 24 and 48 hours. Every 24 hours, serial dilutions of an incubated mixture of fermented porang flour, media, and probiotics were prepared. Then 100µL of the 10^{-5},10^{-6},10^{-7} and 10^{-8} of dilution were spread onto MRS agar and incubated for 48 hours. The viable count of colonies on MRS agar was counted. Only 30-300 colonies were counted.

RESULTS AND DISCUSSION

The pH, total acidity, total bacteria, and yeast of fermented porang's water are shown in Table 1. All treatments showed the same trends, namely a decrease in pH and an increase in total acid during fermentation. The drop in pH during fermentation was also noticed at porang fermentation with starter *A.niger* (Wardhani et al., 2017) and *L.bulgaricus* (Nur'aini et al., 2021). A decrease in pH could be caused by the degradation of glucose into organic acids such as lactic acid and acetic acid by certain microorganisms such as lactic acid bacteria. Both LAB (*L.plantarum*), *B.subtilis,* and *S.cerevisiae* can utilize glucose to become acid (Wang et al., 2021; Mukherjee & Venkata Mohan, 2021; Fakruddin et al., 2017). A decrease in pH was followed by an increase in total acidity. The increase in total acidity was caused by the production of various acids such as acetic acid, lactic acid, and other organic acids (Riesute et al., 2021). The longer the fermentation, the higher the acid was produced. The lowest pH was found in the LP+BS+SC treatment and had the highest total acidity at 72 hours of fermentation. Both *L.plantarum*, *B.subtilis,* and *S.cerevisiae* were able to produce acid so that at 72 hours of fermentation the total acidity and pH in this treatment were higher than in other treatments.

All treatments showed that total aerobic bacteria increased up to 24 hours and decreased from the 48th day of fermentation to the end of fermentation. At the beginning of fermentation, there was still oxygen remaining which allowed aerobic or facultative bacteria to survive and grow up to 48 hours. After 48 hours, there were oxygen depletion and toxic compounds resulting from bacterial and yeast metabolism. These compounds such as lactic acid or other organic acids, ethanol, bacteriocins, hydrogen peroxide, and low pH could inhibit the growth of microorganisms (Joshi et al., 2018; Zang et al., 2020).

The LP+BS and LP+BS+SC treatments had the highest LAB population at 24 hours of fermentation and decreased until the end of fermentation. The addition of LAB to these two treatments before fermentation caused an increase in LAB population at the beginning of fermentation. BS produced bacteriocins which had broad antimicrobial effect than LAB (Caulier et al., 2019). SC produces alcohol and is used as a starter in alcoholic fermentation (Parapouli et al., 2020). The presence of bacteriocin produced by BS and alcohol by SC inhibited the growth of LAB after 48 hours of fermentation. In the LP+SC and BS+SC treatments, the LAB population continued to increase until the end of fermentation. According to Liu et al., (2022) *L. plantarum* cells inhibited *S. cerevisiae* growth with a mechanism when *L. plantarum* cell density reached the threshold at 24 hours, all genes in the lamBDCA quorum sensing (QS) system was upregulated to potentially increase adhesion capability, leading to the aggregation to *S. cerevisiae* cell. This mechanism made the LP population continue to increase until the end of fermentation in the LP+SC treatment. In the BS+SC treatment without the addition of LAB made LAB grew less than in other treatments but the population continued to increase until the end of fermentation. The presence of simple sugars resulting from the breakdown of complex carbohydrates such as starch and glucomannan by

BS bacteria and SC yeast made the LAB population increase until the end of fermentation. It can be seen in Table 2, that the starch content in this treatment is the smallest. It is suspected that the starch had degraded into simple sugars which supported the growth of LAB until the end of fermentation.

All of the treatments had the same dynamic patterns of the yeast population. The yeast population was high at the beginning of fermentation which was derived from raw materials, namely porang tubers. The yeast population decreased at 24 and 48 hours of fermentation. A gradual decrease in yeast was also noticed at cabbage fermentation after 24 hours of fermentation (Satora et al., 2020). At 72 hours of fermentation, the yeast population increased. The decrease in the number of yeasts at 24 and 48 hours was due to the pH not being supportive of yeast growth and competition with aerobic bacteria. Even without the addition of yeast, the LP+BS treatment contained almost the same amount of yeast as the other treatments. Previous studies reported that yeast population during fermentation can be derived from raw material (Kergourlay et al., 2015; Satora et al., 2020). A decrease in the number of yeasts can be caused by conditions not suitable for yeast growth up to 48 hours of fermentation. Yeasts need a low or acidic pH to grow (Maicas, 2020). Some yeast uses organic acids as a carbon source (Satora et al., 2020). The presence of yeast which is tolerant to low pH, and lactic acid increased the increase of yeast population. The high total acid and low pH caused the yeast to increase again at 72 hours of fermentation. According to Rahayu et al. (2021), there is a synergism between yeast, aerobic bacteria, and lactic acid bacteria. During cocoa fermentation, yeast produces vitamins for the growth of LAB and aerobic bacteria. Meanwhile, LAB and acid-producing aerobic bacteria will produce acids that support the growth of yeast.

Table 1. pH, total acidity, and total microorganisms of fermented porang flour.

Treatments	Length of fermentation (hours)	pH	Total acidity (%)	Total aerobic bacteria (log CFU/mL)	Total LAB (log CFU/mL)	Total yeast (log CFU/mL)
LP+BS	0	6.00±0.11	0.1±0.00	5.30±0.75	3.08±0.22	2.96±0.52
	24	5.30±0.03	0.2±0.02	7.90±0.33	5.72±0.15	2.43±0.44
	48	5.06±0.03	0.4±0.03	7.85±0.24	4.49±0.13	2.76±0.31
	72	4.97±0.01	0.6±0.01	6.88±0.97	4.34±0.07	3.29±0.59
LP+SC	0	6.05±0.19	0.1±0.00	5.31±0.12	3.23±0.09	3.38±0.22
	24	5.94±0.05	0.2±0.01	6.73±0.19	4.11±0.15	2.58±0.25
	48	5.07±0.02	0.4±0.00	6.52±0.09	5.59±0.77	2.81±0.16
	72	4.95±0.08	0.7±0.02	5.92±0.17	5.88±0.11	3.82±0.41
BS+SC	0	6.08±0.02	0.1±0.00	6.11±0.81	3.00±0.18	3.28±0.07
	24	5.86±0.06	0.2±0.01	8.05±0.05	3.90±0.02	2.70±0.37
	48	5.17±0.01	0.3±0.01	7.05±0.11	4.60±0.05	2.63±0.07
	72	5.03±0.02	0.5±0.01	5.93±0.18	5.28±0.14	3.41±0.21
LP+BS+SC	0	5.99±0.03	0.1±0.00	4.67±0.23	3.54±0.17	3.19±0.08
	24	5.34±0.07	0.1±0.00	7.59±1.11	6.80±0.23	3.11±0.54
	48	5.22±0.09	0.6±0.02	7.81±0.09	6.77±0.37	3.00±0.25
	72	4.54±0.02	1.2±0.02	6.54±0.05	6.08±0.06	3.89±0.18

At 0 hours of fermentation, it already contained aerobic bacteria, LAB, and yeast. These microorganisms are natural microorganisms that derive from porang tubers. At the beginning of fermentation, it was assumed that yeast used simple glucose to produce ethanol. Ethanol can be converted into acetic acid by aerobic bacteria (Rahayu et al., 2021). At 24 hours, the available glucose was used by BAL to produce lactic acid. During fermentation, all treatments showed that aerobic bacteria and LAB played a role at 24 to 48 hours of fermentation, while yeast at 72 hours of fermentation. The growth of bacteria at the beginning of fermentation, especially BS and LP, played a role in degrading complex carbohydrates such as starch into simple glucose. Yeast uses simple glucose to convert it into ethanol (Helmi et al., 2024). At the end of fermentation, bacteria decreased which was caused by the alcohol and acid content. In line with Ambonese arrack

fermentation and inasua traditional fermented fish, the number of microorganisms decreased at the end of fermentation due to the accumulation of alcohol and acid content because fermentation took place in batch culture (Mahulette et al., 2018; Mahulette & Astuti, 2020).

The fermentation process caused an increase in the glucomannan content of porang tubers compared to without fermentation (Table 2). However, in the LP+BS+SC treatment, the glucomannan content was smaller than without fermentation. It could be caused by glucomannan that had been degraded into glucose and mannan at 72 hours of fermentation. In line with pH and total acid at 72 hours of fermentation (Table 1), the LP+BS+SC treatment had the lowest pH and the highest total acid (acidity). The lowest pH and the highest acidity were caused by the degradation of glucose to organic acids. Moreover, LP+BS+SC also had the lowest starch content. It was assumed that the low level of starch was caused by the degradation of starch into simpler molecules such as glucose. Glucose can be converted into organic acids such as acetic acid and lactic acid or converted into alcohol (Joshi et al., 2018). These results showed that 72 hours was not the right time for stopping fermentation for the LP+BS+SC treatment.

The BS+SC treatment had the highest glucomannan levels. This treatment also has the lowest starch content and the highest carbohydrate content. In this treatment, the high carbohydrate content was derived from glucomannan. Starch is one of the impurities in glucomannan flour. The results showed that at 72 hours of fermentation, BS+SC treatment was the best treatment to produce high glucomannan flour with few impurities. This treatment also contained higher fat and lower protein. In addition to the BS+SC treatment, it had the lowest protein content, even lower than without fermentation. BS has protease activity (Soeka & Sulistiani, 2014) as well as SC. It meant that the protein in the BS+SC fermented flour at 72 hours of fermentation had been degraded.

Table 2. The proximate, crude fiber, starch, and glucomannan content of non-fermented and fermented porang flour after 72 days of fermentation.

Parameter	Non Fermented Porang Flour	LP+BS	LP+SC	BS+SC	LP+BS+SC
Water (%)	10.70±0.13	11.26±0.13	10.80±0.08	10.52±0.16	10.70±0,13
Ash (%)	6.08±0.06	5.82±0.04	5.14±0.02	5.05±0.13	5.16±0.15
Protein (%)	8.03±0.18	8.48±0.23	7.47±0.17	7.25±0.16	8.03±0.11
Fat (%)	1.59±0.05	1.73± 0.04	1.64±0.04	1.86±0.05	1.24±0.04
Carbohydrate (%)	73.62±0.47	72.72±0.43	74.96±0.27	75.33±0.08	74.82±0.47
Starch (%)	51.46±0.74	52.60±0.08	54.29±0.82	49.93±0.79	48.00±0.16
Fiber (%)	1.13±0.02	3.10±0.11	1.85±0.04	4.50±0.09	3.38±0.09
Glucomannan (%)	28.90±4.79	47.23±7.61	61.14±3.81	65.73±4.88	15.04±1.19

Table 2 shows that the fermented porang flour with the treatment BS+SC had higher fiber content. This could be caused by the degradation of the fibers into fine fibers so that they pass through the 90-mesh sieve. In the treatment without BS, the fiber had not been degraded so it does not pass through the 90-mesh sieve so it has a lower fiber content. BS is a bacteria that can dissolve fiber through cellulolytic activity (Kresnowati et al., 2019).

Table 2 shows that the LP+BS treatment had the highest water content. The highest ash content was reached by flour without fermentation. It was caused by the high content of calcium oxalate in unfermented porang. Porang belongs to the Aracaceae family (Kumoro et al., 2019) and contains calcium oxalate (Chairiyah et al., 2016). Porang fermented flour with BS+SC treatment had the lowest calcium oxalate. It could be caused by high levels of organic acids which was produced by BS+SC. Organic acids can dissolve the calcium oxalate found in porang (Agustin et al., 2017; Wardani & Arifiyana, 2020). BS produces various organic acids and can convert alcohol into various acids. BS can produce lactate, acetate, butaniol, and a small amount of alcohol (Mukherjee & Venkata Mohan, 2021). *Saccharomyces cerevisiae* produced ethanol (Varize et al., 2022). Aerobic bacteria such as BS could change ethanol to acetic acid (Rahayu et al., 2021).

Figure 1 shows the antibacterial activity of fermented porang flour after 72 hours of fermentation against gram-positive and negative bacteria. Antibacterial test results showed that

BS+SC treatment had antibacterial activity which began at a concentration of 10%. Porang flour with BS+SC treatment was able to inhibit *E. coli* and *S.aureus*. Metabolites which were produced by bacteria, namely various acids and alcohol imbibed into the flour. BS had activity to inhibit *Staphylococcus aureus, Escherichia coli,* and *Pseudomonas aeruginosa* (Singh et al., 2018). SC also has antibacterial activity against both Gram-positive and Gram-negative bacteria (Al-sahlany et al., 2020). SC produced peptides with a molecular weight of 99.7 kilodaltons with a pH of 4-7 to inhibit *E.coli* and *S.aureus* with the adsorption mechanism of bioactive peptides. These peptides degrade bacterial cell membranes, leading to complete damage to the membrane (Al-sahlany et al., 2020). SC isolated from fruit has antibacterial, antifungal, antioxidant properties and potential probiotics (Fakruddin et al., 2017). Yeast can also produce killer toxins to inhibit other microorganisms (Satora et al., 2020). Based on Table 3, fermented porang flour inhibited Gram-positive bacteria more than Gram-negative bacteria. According to Al-sahlany et al. (2020) and Makky et al. (2021), *S.cerevisiae* inhibited more Gram-negative bacteria than Gram-positive bacteria. The presence of secondary metabolites produced by *B. subtilis* was assumed to influence the inhibition mechanism of fermented porang flour against bacteria. *B.subtilis* produces various secondary metabolites that can inhibit other bacteria (Caulier et al., 2019). BS had bacteriocins with a broader spectrum than *Lactobacillus*. The mixture of metabolites from BS and SC changed the bacterial inhibitory mechanism when compared to the metabolites produced by either microorganism alone.

Table 3. The antibacterial activity of fermented porang flour against *E.coli* and *S.aureus*

Bacteria	Concentration (%)	Inhibition zone (mm)			
		LP+BS	LP+SC	BS+SC	LP+BS+SC
Escherichia coli	Positive control	0	0	46.03±8.30	0
	1	0	0	0	0
	10	0	0	7.07±1.01	0
	50	0	0	16.10±5.95	0
Staphylococcus aureus	Positive control	0	0	42.41±2.58	0
	1	0	0	0	0
	10	0	0	8.55±1.82	0
	50	0	0	24.62±2.79	0

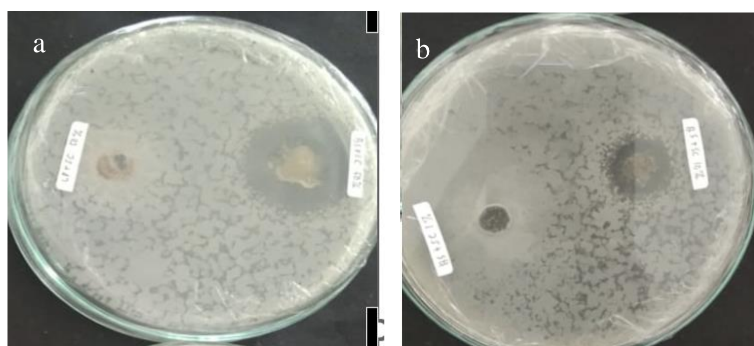

Figure 1. The clear (inhibition) zone of fermented porang flour against *S.aureus*. (a). Inhibition zone of porang flour with addition of LP+SC (left), and BS+SC (right) at 50% concentration, respectively; (b). Inhibition zone of fermented porang flour with addition BS+SC at 1% concentration (left) and 10% concentration (right).

Prebiotics showed that fermented porang's flour may be developed to be prebiotics (Table 4). The porang fermented flour could increase the population of *L. acidophilus* except for the fermented flour with treatment LP+BS+SC. In line with glucomannan content in Table 2, LP+BS+SC contained the lowest glucomannan content. Glucomannan is known as Generally recommended as safe (GRAS) so it is safe to consume (Zhou et al., 2022). Glucomannan was known as a prebiotic. The prebiotic ability of glucomannan was caused by KGM gels can

provide a continuously moist environment. KGM has properties to regulate the stability and rheological properties of emulsions, which provides an interesting prospect for the development of prebiotic emulsions with multiple applications (Zhou et al., 2022). Isolate *Bacillus subtilis* which was isolated from Hentak (India 's fermented food), had high antimicrobial activity against *Staphylococcus aureus*, *Escherichia coli,* and *Pseudomonas aeruginosa*. Moreover, this bacterium had probiotic activity due to the capability to form biofilm and survive at low pH (4.0) (Singh et al., 2018). *S.cereviase* is also known as a probiotic (Fakruddin et al., 2017). *S.cereviseae* produces some metabolite compounds, namely organic acids as well as vitamin B. *S.cereviase* can assimilate cholesterol and produce killer toxins (Fakruddin et al., 2017). *L.acidophilus* is a probiotic bacterium because this bacterium is resistant to bile acid and gastric juice. *L.acidophilus* has antioxidant ability and protective effects against cell damage (Hoffmann et al., 2019).

Table 4. The prebiotic activity of fermented porang flour to *L.acidophilus*

Fermented porang flour	Total colony (log CFU/gram)	
	24 hours	48 hours
Non Fermented Porang Flour	7.34±0.34	8.60±0.02
LP+BS	7.43±0.58	8.88±0.23
LP+SC	8.33±0.06	9.16±0.20
BS+SC	8.63±0.63	8.89±0.08
LP+BS+SC	6.88±0.22	8.14±0.47

Overall, the addition of a starter may have benefits for the industry of porang flour. Fermented porang flour can increase the nutritional and functional value of porang flour. The addition of a starter can reduce the anti-nutritional compound in porang flour, namely calcium oxalate, which can be noticed from the decrease in ash content. The use of starters, namely *B.subtilis* and *S.cerevisiae* can effectively increase glucomannan and carbohydrate levels. Porang fermented flour produced by adding these bacteria can improve the purity of glucomannan, as seen from the low starch content. Porang flour is used in the food industry and also as a diet for weight loss. The highest fiber content of porang fermented flour with the addition of *B.subtilis* and *S.cerevisiae* may suit for diet. Fermented porang flour with the addition of *B. subtilis* and *S. cerevisiae* also had the best antibacterial and prebiotic capabilities. It is hoped that the use of these bacteria as starters in the porang tuber processing industry can provide added value and increase Indonesian porang exports.

CONCLUSION

Fermentation can increase the glucomannan levels of porang tubers. Fermented porang flour with adding *Bacillus subtilis* and *Saccharomyces cerevisiae* at 72 hours of fermentation can be developed as a nutritional and functional food because it contains high glucomannan, has antibacterial activity, and has prebiotic properties. Moreover, this treatment also had the highest fiber and carbohydrate content. The pH and total acidity at 72 hours of fermentation of porang tubers with the treatment of *Lactobacillus plantarum*, *Bacillus subtilis,* and *Saccharomyces cerevisiae* showed that carbohydrate and glucomannan had been degraded faster than other treatments so that it had the lowest glucomannan and starch content. In future research, the study about the dynamics of organic acids and ethanol content during porang fermentation using *B.subtilis* and *S.cerevisiae* starters needs to be observed to better understand the fermentation mechanism or process so that the quality of the porang flour produced can be improved.

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REFERENCES

Afzaal, M., Saeed, F., Islam, F., Ateeq, H., Asghar, A., Shah, Y. A., Ofoedu, C. E., & Chacha, J. S. (2022). Nutritional Health Perspective of Natto: A Critical Review. *Biochemistry Research*

International, *2022*. https://doi.org/10.1155/2022/5863887

Agustin, R., Estiasih, T., & Wardani, A. (2017). Decrease of Oxalate on Construction Process of New Cocoyam (Xanthosoma Sagittifolium) in Various Consentration of Acetic Acid. *Jurnal Teknologi Pertanian*, *18*(3), 191–200. https://doi.org/10.21776/ub.jtp.2017.018.03.19

Al-sahlany, S. T. G., Altemimi, A. B., Abd Al-Manhel, A. J., Niamah, A. K., Lakhssassi, N., & Ibrahim, S. A. (2020). Purification of Bioactive Peptide with Antimicrobial Properties Produced by Saccharomyces cerevisiae. *Foods*, *9*(3), 1–11. https://doi.org/10.3390/foods9030324

Alika, V. A., & Atma, Y. (2018). The organoleptic and physicochemical characteristic of gelato by fish bone gelatin addition. *Journal of Applied Science and Advanced Technology*, *1*(1), 31–38.

Andriani, Y., Safitri, R., Rochima, E., & Fakhrudin, S. D. (2017). Characterization of Bacillus subtilis and B. licheniformis potentials as probiotic bacteria in Vanamei shrimp feed (Litopenaeus vannamei Boone, 1931). *Nusantara Bioscience*, *9*(2), 188–193. https://doi.org/10.13057/nusbiosci/n090214

Anggela, Harmayani, E., Setyaningsih, W., & Wichienchot, S. (2022). Prebiotic effect of porang oligo-glucomannan using fecal batch culture fermentation. *Food Science and Technology (Brazil)*, *42*, 1–7. https://doi.org/10.1590/fst.06321

AOAC. (2006). *Official methods of analysis* (18th ed.). AOAC International.

Apriliani, A. L., & Mulyadi, A. H. (2022). Analysis of Characteristics of Mocaf (Modified Cassava Flour) with Variations of Fermentation Time and Bread Yeast Concentration (Saccharomyces cerevisiae). *Research In Chemical Engineering (RiCE)*, *1*(2), 59–63. https://doi.org/10.30595/rice.v1i2.26

Behera, S. S., & Ray, R. C. (2016). Konjac glucomannan, a promising polysaccharide of Amorphophallus konjac K. Koch in health care. *International Journal of Biological Macromolecules*, *92*, 942–956. https://doi.org/10.1016/j.ijbiomac.2016.07.098

Budiastra, I. W., & Noviyanti, A. A. (2023). Determination of Chemical Content of Porang Flour (Amorphophallus muelleri blume) by Near Infrared Spectroscopy. *IOP Conference Series: Earth and Environmental Science*, *1187*(1). https://doi.org/10.1088/1755-1315/1187/1/012027

Caulier, S., Nannan, C., Gillis, A., Licciardi, F., Bragard, C., & Mahillon, J. (2019). Overview of the antimicrobial compounds produced by members of the Bacillus subtilis group. *Frontiers in Microbiology*, *10*(FEB), 1–19. https://doi.org/10.3389/fmicb.2019.00302

Chairiyah, N., Harijati, N., & Mastuti, R. (2016). Variation of Calcium Oxalate (CaOx) Crystals in Porang Corms (<i>Amorphophallus muelleri</i> Blume) at Different Harvest Time. *American Journal of Plant Sciences*, *07*(02), 306–315. https://doi.org/10.4236/ajps.2016.72030

Chua, M., Chan, K., Hocking, T. J., Williams, P. A., Perry, C. J., & Baldwin, T. C. (2012). Methodologies for the extraction and analysis of konjac glucomannan from corms of Amorphophallus konjac K. Koch. *Carbohydrate Polymers*, *87*(3), 2202–2210. https://doi.org/10.1016/j.carbpol.2011.10.053

Cuamatzin-garcía, L., Rodríguez-rugarcía, P., El-kassis, E. G., Galicia, G., Meza-jiménez, M. de L., Baños-lara, M. D. R., Zaragoza-maldonado, D. S., & Pérez-armendáriz, B. (2022). Traditional Fermented Foods and Beverages from around the World and Their Health Benefits. *Microorganisms*, *10*(6), 1–18. https://doi.org/10.3390/microorganisms10061151

David, S., Femi, B., Gbenga, A., & Ab, S. (2018). Purification and Characterization of α -Amylase from Bacillus subtilis Isolated from Cassava Processing Sites. *Journal of Bioremediation & Biodegradation Purification*, *8*(6), 1–7. https://doi.org/10.4172/2155-6199.1000417

Devaraj, R. D., Reddy, C. K., & Xu, B. (2019). Health-promoting effects of konjac glucomannan and its practical applications: A critical review. *International Journal of Biological Macromolecules*, *126*, 273–281. https://doi.org/10.1016/j.ijbiomac.2018.12.203

Dimidi, E., Cox, S., Rossi, M., & Whelan, K. (2019). Fermented Foods: Definitions and Characteristics , Gastrointestinal Health and Disease. *Nutrients*, *11*(1806), 1–26.

Dwiloka, B., Setiani, B. E., & Irsalina, K. (2019). Moisture Content , Protein , Crude Fiber and Antioxidant Activity of Cookies with Boiled Papaya Leaf. *IOP Conference Series: Earth and Environmental Science*, *292*(012044). https://doi.org/10.1088/1755-1315/292/1/012044

Fakruddin, M., Hossain, M. N., & Ahmed, M. M. (2017). Antimicrobial and antioxidant activities of Saccharomyces cerevisiae IFST062013, a potential probiotic. *BMC Complementary and Alternative Medicine*, *17*(1), 1–11. https://

doi.org/10.1186/s12906-017-1591-9

Feng, R., Chen, L., & Chen, K. (2018). Fermentation trip: amazing microbes, amazing metabolisms. *Annals of Microbiology*, *68*(11), 717–729. https://doi.org/10.1007/s13213-018-1384-5

Fidanza, M., Panigrahi, P., & Kollmann, T. R. (2021). Lactiplantibacillus plantarum–Nomad and Ideal Probiotic. *Frontiers in Microbiology*, *12*(October), 1–13. https://doi.org/10.3389/fmicb.2021.712236

Gómez, B., Míguez, B., Yáñez, R., & Alonso, J. L. (2017). Manufacture and Properties of Glucomannans and Glucomannooligosaccharides Derived from Konjac and Other Sources. *Journal of Agricultural and Food Chemistry*, *65*(10), 2019–2031. https://doi.org/10.1021/acs.jafc.6b05409

Harmayani, E., Aprilia, V., & Marsono, Y. (2014). Characterization of glucomannan from Amorphophallus oncophyllus and its prebiotic activity in vivo. *Carbohydrate Polymers*, *112*, 475–479. https://doi.org/10.1016/j.carbpol.2014.06.019

Hattingh, M., Alexander, A., Meijering, I., Van, R. C. A., & Dicks, L. M. T. (2015). Amylolytic strains of Lactobacillus plantarum isolated from barley. *African Journal of Biotechnology*, *14*(4), 310–318. https://doi.org/10.5897/ajb2014.14149

Helmi, H., Kusmiadi, R., Mahardika, R. G., & Karsiningsih, E. (2024). Diversity of fungi and dynamics of ethanol concentration during fermentation of porang (Amorphophalus oncophyllus). *Biodiversitas*, *25*(2), 542–552. doi: 10.13057/biodiv/d250213

Helmi, H., Susanti, I., Agung, N. A., & Kusen, S. (2015). Antibacterial activity of belilik (Brucea javanica (L). MERR) growth of enteropathogenic bacteria. *Journal of Biological Research*, *21*(1), 35–40.

Hoffmann, A., Kleniewska, P., & Pawliczak, R. (2019). State of the art paper Antioxidative activity of probiotics. *Nature Reviews Neuroscience*, 792–804. https://doi.org/10.5114/aoms.2019.89894

Hu, Q., & Liu, J. (2021). Production of α-Amylase by Bacillus subtilis QM3 and its Enzymatic Properties. *OALib*, *08*(03), 1–8. https://doi.org/10.4236/oalib.1107291

Joshi, R., Sharma, V., & Kuila, A. (2018). Fermentation Technology: Current Status and Future Prospects. In *Principles and Applications of Fermentation Technology* (Issue August). https://doi.org/10.1002/9781119460381.ch1

Kaminarides, S., Moschopoulou, E., & Karali, F. (2019). Influence of salting method on the chemical and texture characteristics of ovine Halloumi cheese. *Foods*, *8*(7). https://doi.org/10.3390/foods8070232

Kergourlay, G., Taminiau, B., Daube, G., & Champomier Vergès, M. C. (2015). Metagenomic insights into the dynamics of microbial communities in food. *International Journal of Food Microbiology*, *213*, 31–39. https://doi.org/10.1016/j.ijfoodmicro.2015.09.010

Kresnowati, M., Turyanto, L., Zaenuddin, A., & Trihatmoko, K. (2019). Effects of microbial starter composition on nutritional contents and pasting properties of fermented cassava flour. *ASEAN Journal of Chemical Engineering*, *19*(1), 12–24. https://doi.org/10.22146/ajche.50871

Kumar, V., Sheoran, P., Gupta, A., Yadav, J. P., & Tiwari, S. K. (2016). Antibacterial property of bacteriocin produced by Lactobacillus plantarum LD4 isolated from a fermented food. *Annals of Microbiology*, *66*(4), 1431–1440. https://doi.org/10.1007/s13213-016-1230-6

Kumoro, A. C., Amyranti, M., Retnowati, D. S., & Ratnawati, R. (2019). Browning Prevention of Chips from Freshly Harvested Porang (Amorphophallus oncophyllus) Tubers through Immersion in Ascorbic Acid Solutions at Various Times. *Journal of Physics: Conference Series*, *1295*(1), 1–6. https://doi.org/10.1088/1742-6596/1295/1/012023

Liu, J., Huang, T.-Y., Liu, G., Ye, Y., Soteyome, T., Seneviratne, G., Xiao, G., Xu, Z., & Kjellerup, B. V. (2022). Microbial Interaction between Lactiplantibacillus plantarum and Saccharomyces cerevisiae: Transcriptome Level Mechanism of Cell-Cell Antagonism. *Microbiology Spectrum*, *10*(5), 1–18. https://doi.org/10.1128/spectrum.01433-22

Mahulette, F., & Astuti, D. I. (2020). Microbial Succession and Chemical Characteristics in Fermentation of Ambonese arrack (Sopi), Traditional Beverage from Maluku. *Biosaintifika*, *12*(2), 147–154. https://doi.org/10.15294/biosaintifika.v12i2.23791

Mahulette, F., Mubarik, N. R., Suwanto, A., & Widanarni. (2018). Microbiological and Physicochemical Characteristics of Inasua Traditional Fish Fermented from Maluku Islands. *Biosaintifika*, *10*(2), 298–305. https://doi.org/10.15294/biosaintifika.v10i2.13537

Maicas, S. (2020). The role of yeasts in fermentation

processes. *Microorganisms*, *8*(8), 1–8. https://doi.org/10.3390/microorganisms8081142

Makky, E. A., Almatar, M., Mahmood, M. H., Wei Ting, O., & Zi Qi, W. (2021). Evaluation of the antioxidant and antimicrobial activities of ethyl acetate extract of saccharomyces cerevisiae. *Food Technology and Biotechnology*, *59*(2), 127–136. https://doi.org/10.17113/ftb.59.02.21.6658

Mani, A. (2018). Food Preservation by Fermentation and Fermented Food Products. *International Journal of Academic Research & Development*, *1*, 51–57. https://www.researchgate.net/publication/323200130

Mukherjee, T., & Venkata Mohan, S. (2021). Metabolic flux of Bacillus subtilis under poised potential in electrofermentation system: Gene expression vs product formation. *Bioresource Technology*, *342*(August), 125854. https://doi.org/10.1016/j.biortech.2021.125854

Nur'aini, Dari, I. W., Setiawan, A. A., & Laksmitawati, D. R. (2021). Macronutrients Analysis of Porang Tubers (Amorphophallus muelleri Blume) Fermentation With Lactobacillus Bulgaricus Bacteria . *Proceedings of the 4th International Conference on Sustainable Innovation 2020–Health Science and Nursing (ICoSIHSN 2020)*, *33*(ICoSIHSN 2020), 488–492. https://doi.org/10.2991/ahsr.k.210115.096

Nurlela, Ariesta, N., Laksono, D. S., Santosa, E., & Muhandri, T. (2021). Characterization of glucomannan extracted from fresh porang tubers using ethanol technical grade. *Molekul*, *16*(1), 1–8. https://doi.org/10.20884/1.jm.2021.16.1.632

Nurlela, Ariesta, N., Santosa, E., & Muhandri, T. (2019). Effect of harvest timing and length of storage time on glucomannan content in porang tubers. *IOP Conference Series: Earth and Environmental Science*, *299*(1). https://doi.org/10.1088/1755-1315/299/1/012012

Parapouli, M., Vasileiadis, A., Afendra, A. S., & Hatziloukas, E. (2020). Saccharomyces cerevisiae and its industrial applications. *AIMS Microbiology*, *6*(1), 1–31. https://doi.org/10.3934/microbiol.2020001

Penido, F. C. L., Piló, F. B., Sandes, S. H. de C., Nunes, Á. C., Colen, G., Oliveira, E. de S., Rosa, C. A., & Lacerda, I. C. A. (2018). Selection of starter cultures for the production of sour cassava starch in a pilot-scale fermentation process. *Brazilian Journal of Microbiology*, *49*(4), 823–831. https://doi.org/10.1016/j.bjm.2018.02.001

Persson, J. A., Wennerholm, M., & O'Halloran, S. (2008). Handbook for Kjeldahl Digestion. In *FOSS, Hilleroed, Denmark*. FOSS, Hilleroed, Denmark. https://doi.org/10.3168/jds.2009-2906

Rahayu, E. S., Triyadi, R., Khusna, R. N. B., Djaafar, T. F., Utami, T., Marwati, T., & Hatmi, R. U. (2021). Indigenous yeast, lactic acid bacteria, and acetic acid bacteria from cocoa bean fermentation in indonesia can inhibit fungal-growth-producing mycotoxins. *Fermentation*, *7*(3), 2–11. https://doi.org/10.3390/fermentation7030192

Raul, D., Biswas, T., Mukhopadhyay, S., Das, S. K., & Gupta, S. (2014). *Production and Partial Purification of Alpha Amylase from Bacillus subtilis (MTCC 121) Using Solid State Fermentation. 2014*.

Riesute, R., Salomskiene, J., Moreno, D. S., & Gustiene, S. (2021). Effect of yeasts on food quality and safety and possibilities of their inhibition. *Trends in Food Science and Technology*, *108*, 1–10. https://doi.org/10.1016/j.tifs.2020.11.022

Satora, P., Skotniczny, M., Strnad, S., & Ženišová, K. (2020). Yeast microbiota during sauerkraut fermentation and its characteristics. *International Journal of Molecular Sciences*, *21*(24), 1–15. https://doi.org/10.3390/ijms21249699

Sharma, R., Garg, P., Kumar, P., Bhatia, S. K., & Kulshrestha, S. (2020). Microbial fermentation and its role in quality improvement of fermented foods. *Fermentation*, *6*(4), 1–20. https://doi.org/10.3390/fermentation6040106

Singh, S. S., De Mandal, S., Lalnunmawii, E., & Senthil Kumar, N. (2018). Antimicrobial, antioxidant and probiotics characterization of dominant bacterial isolates from traditional fermented fish of Manipur, North-East India. *Journal of Food Science and Technology*, *55*(5), 1870–1879. https://doi.org/10.1007/s13197-018-3103-4

Song, E., Ang, L., Lee, H. W., Kim, M. S., Kim, Y. J., Jang, D., & Lee, M. S. (2023). Effects of kimchi on human health: a scoping review of randomized controlled trials. *Journal of Ethnic Foods*, *10*(1). https://doi.org/10.1186/s42779-023-00173-8

Suryana, E. A., Kamsiati, E., Usmiati, S., & Herawati, H. (2022). Effect of Porang Flour and Low-Calorie Sugar Concentration on the Physico-Chemical Characteristics of Jelly Drinks. *IOP Conference Series: Earth and Environmental Science*, *985*(1). https://doi.org/

10.1088/1755-1315/985/1/012042

Varize, C. S., Bücker, A., Lopes, L. D., Christofoleti-Furlan, R. M., Raposo, M. S., Basso, L. C., & Stambuk, B. U. (2022). Increasing Ethanol Tolerance and Ethanol Production in an Industrial Fuel Ethanol Saccharomyces cerevisiae Strain. *Fermentation*, *8*(10), 1–14. https://doi.org/10.3390/fermentation8100470

Wahidah, B. F., Afiati, N., & Jumari. (2021). Community knowledge of amorphophallus muelleri blume: Cultivation and utilization in central java, indonesia. *Biodiversitas*, *22*(7), 2731–2738. https://doi.org/10.13057/biodiv/d220722

Wang, Y., Wu, J., Lv, M., Shao, Z., Hungwe, M., Wang, J., Bai, X., Xie, J., Wang, Y., & Geng, W. (2021). Metabolism Characteristics of Lactic Acid Bacteria and the Expanding Applications in Food Industry. *Frontiers in Bioengineering and Biotechnology*, *9*(May), 1–19. https://doi.org/10.3389/fbioe.2021.612285

Wardani, R. K., & Arifiyana, D. (2020). The Effect of Soaking Time and Temperature of Acetic Acid Solution to the De-crease of Calcium Oxalate Levels in Porang Tubers. *Nst Proceedigs*, *2020*, 145–149. https://doi.org/10.11594/nstp.2020.0522

Wardhani, D. H., Aryanti, N., Haryani, K., & Maharani, F. (2017). Kinetic study of solid fermentation of Amorphophallus oncophyllus flour using Aspergillus niger And Its Prebiotic potential. *Carpathian Journal of Food Science and Technology*, *9*(2), 43–56.

Wardhani, D. H., Vázquez, J. A., Ramdani, D. A., Lutfiati, A., Aryanti, N., & Cahyono, H. (2019). Enzymatic purification of glucomannan from amorphophallus oncophyllus using A-amylase. *Bioscience Journal*, *35*(1), 277–288. https://doi.org/10.14393/BJ-v35n1a2019-41766

Wewo, N. M., Prasetyawan, S., & Srihardyastutie, A. (2018). Antinutritional Content, Flour Characteristics, and Protein Profiles of Taro Tubers (Colocasia esculenta) by Fermentation with Lactobacillus plantarum. *The Journal of Pure and Applied Chemistry Research*, *7*(3), 301–307. https://doi.org/10.21776/ub.jpacr.2018.007.03.425

Widjanarko, S. B., Affandi, M., & Wahyuli, Z. (2022). A review on konjac glucomannan and hydrolysed konjac glucomannan. *Food Research*, *6*(5), 425–433. https://doi.org/10.26656/fr.2017.6(5).920

Zang, J., Xu, Y., Xia, W., & Regenstein, J. M. (2020). Quality, functionality, and microbiology of fermented fish: a review. *Critical Reviews in Food Science and Nutrition*, *60*(7), 1228–1242. https://doi.org/10.1080/10408398.2019.1565491

Zhao, C. J., Schieber, A., & Gänzle, M. G. (2016). Formation of taste-active amino acids, amino acid derivatives and peptides in food fermentations – A review. *Food Research International*, *89*, 39–47. https://doi.org/10.1016/j.foodres.2016.08.042

Zhou, N., Zheng, S., Xie, W., Cao, G., Wang, L., & Pang, J. (2022). Konjac glucomannan: A review of structure, physicochemical properties, and wound dressing applications. *Journal of Applied Polymer Science*, *139*(11), 1–16. https://doi.org/10.1002/app.51780

Zhu, L., Jones, C., Guo, Q., Lewis, L., Stark, C. R., & Alavi, S. (2016). An evaluation of total starch and starch gelatinization methodologies in pelleted animal feed. *Journal of Animal Science*, *94*(4), 1501–1507. https://doi.org/10.2527/jas.2015-9822

Zubaidah, E., Susanti, I., Yuwono, S. S., Rahayu, A. P., Srianta, I., & Blanc, P. J. (2020). Effect of lactobacillus plantarum and leuconostoc mesenteroides starter cultures in lower salt concentration fermentation on the sauerkraut quality. *Food Research*, *4*(4), 1038–1044. https://doi.org/10.26656/fr.2017.4(4).029