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Lactic acid fermentation of banana peel using *Lactobacillus plantarum* : Effect of substrate concentration, inoculum concentration, and various nitrogen sources

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Abstract

Agung Semeru Banana peel is an organic waste that is exclusively utilized as animal feed and does not harm the environment. The primary component of banana peels is carbohydrates, which can be used as a substrate during the fermentation process to produce lactic acid. The fermentation of banana peel flour with Lactobacillus plantarum strain FNCC 0020 was the main focus of this investigation. Variations in the concentrations of the substrate and inoculum as well as the impact of the type of nitrogen on lactic acid concentration were investigated. According to research findings, the big banana peel contains 70.52% carbs, 5.68% soluble protein, 3.115% fat, 6.74% water, 2.395% ash, and 13.38% crude fiber. While the inoculum variable was 0.5% v/v and the best substrate concentration variable was 17.5% w/v, the best lactic acid concentrations were 5.401 g/L and 8.586 g/L, respectively, as determined by HPLC (High-Performance Liquid) analysis. Banana peel flour only includes a modest amount of nitrogen (0.8295%), sulfate (0.037 grams), phosphate (1.6105%), and vitamin B1 (0.2315%), so additional nitrogen sources must be added. The production of lactic acid is shown to increase with the addition of various forms of nitrogen, with ammonium sulfate and ammonium phosphate (2:1) producing the greatest yields of 9.781 g/L and 14.255 g/L, respectively, of lactic acid, which is lower than lactic acid from yeast extract.

Keywords: lactic acid; fermentation; banana peels; *lactobacillus plantarum*

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INTRODUCTION

Carl Wilhelm Scheele, a Swedish chemical scientist, made the initial discovery in 1780 while trying to remove acid lactic from condensed milk. A acidic early organic is acid lactic (LA), also referred to as 2-hydrocyanic acid. Pasteurization technology was already in use in 1857, but sour lactic acid fermentation was still a relatively new occurrence. Continue to be examined due to the potential involvement of microbes. French Fre, an innovative scientist, introduced acid lactic, the first bacteria utilized in the United States, in 1881 (Ghaffar et al., 2014). From 1220.0 kilotons in 2016 to 1960.1 kilotons in 2025, the global demand for lactic acid will continue to rise (Alves de Oliveira et al., 2018). It is predicted that from 2022 to 2030, the market demand, which was worth USD 2.9 billion in 2021, will increase at a compound annual growth rate (CAGR) of 8.0%. Numerous sectors, particularly those in developing nations, have a substantial market for the usage of lactic acid. It is predicted that developing nations such as India, China, and Indonesia will be the main drivers of demand for these items in the future (Mordor Intelligence, 2017). As a result, there is the potential for the use of lactic acid to increase by 5 to 8% annually, and is pervasive in many industrial sectors, including food, chemical, cosmetic, and other industries (Yadav et al., 2011). The presence of carboxyl and hydroxyl groups in lactic acid's structure allows it to be transformed into a variety of acids, esters, and bio-solvents that can be employed as flavors, bacterial inhibitors, and important roles in many fields (Rodrigues et al., 2016). The fact that lactic acid may be utilized as a raw material to create environmentally friendly, biodegradable PLA (*Poly Lactic Acid*) provides an even more exciting prospect for the substance (Tian et al., 2018). A green polymer that can replace the use of plastics made from petroleum is called PLA, which is produced via the lactic acid polymerization process (Batista da Mota et al., 2022). In the medical field, bioabsorbable PLA is employed as a biomaterial that regenerates tissue healing, sutures, and implants in addition to its role as a replacement for plastics such as rubbish bags, tarpaulins, agricultural plastics, and food packaging (Hiraishi, 2010; Mochizuki, 2010; Obuchi & Ogawa, 2010; Suzuki & Ikada, 2010).

Research on the manufacture of lactic acid from renewable substrates, including milk waste, cellulose from agriculture and forestry waste, and food waste, including wheat bran, maize steep liquid, and molasses, has been done (Abedi & Hashemi, 2020; Ghaffar et al., 2014; Hwang et al., 2012). Banana peels can be used to create various renewable substrates. India is the world's biggest banana producer, followed by China, Indonesia, Brazil, and Ecuador also produce significant amounts of bananas, which are largely farmed in Asia, Latin America, and Africa (FAOSTAT, 2020). The production of banana fruit commodities climbed 11.281% from the previous year, with a total banana product of almost 1,231,218 quintals in 2021. Lumajang is one of the Indonesian towns that produce the most split fruit in East Java, with a total product from 21 sub-districts in Lumajang Regency. According to information from the Department of Industry and Trade of the Lumajang Regency, which in 2013 counted 5,428 industries, 197 industries (3.63%) of the entire industry engaged in the processing of giant bananas into different foods (Budiwati, 2016). In Lumajang Regency, there are up to 430,926.3 quintals of banana peels produced every year based on the fact that 35% of bananas peels. Banana peels have the potential to be utilized for other purposes besides animal feed and are currently solely used as waste. The steps in the production of lactic acid include the breakdown of carbohydrates into monosaccharides and the use of enzymes made by lactic acid bacteria that will turn the monosaccharides into lactic acid. A quality of *Lactobacillus plantarum* is its ability to withstand high acidity, which enables the fermentation process to proceed almost contamination free. People can use the homofermentative *Lactobacillus plantarum* (LAB) to ferment lactose into lactic acid. This process is advantageous because it not only uses protein contained in the substrate but also utilizes additional nutrients, such as lactose, at a high conversion rate (Fu & Mathews, 1999). Due to the homofermentative nature of Lactobacillus plantarum, which produces L(+) lactic acid, it is widely used to improve the quality of fermentation (Dharmalingam et al., 2012; Ray et al., 2009), (Wang et al., 2020; Xu et al., 2022). Polylactic acid (PLA) can be successfully produced using a variety of substrates, as demonstrated by (Helmes et al., 2018; Mekonnen et al., 2013; Tsapekos et al., 2020). Banana peels from the Enterococcus faecium FW26 strain were used in a study that resulted in fermentation pH control reaching 33.32.6g/L and lactic acid production of 15.9g/L. The thermo alkali lactic acid bacteria *Enterococcus durans* BP130 from banana peels (BP) produced the most lactic acid during batch fermentation when $CaCO_3$ was used as a neutralizer. (Hassan et al., 2019).

Agung Semeru banana peel flour was used in this experiment as a substrate for the batch fermentation of *Lactobacillus plantarum* strain FNCC 0020. The price of raw materials, which includes the cost of manufacturing lactic acid, is the first factor that becomes important. In addition to the inexpensive substrate, other sources are used to encourage the production of lactic acids, such as the addition of nitrogen sources during the process.

RESEARCH METHOD

Producing Agung Semeru banana peel flour

Banana peel from an Agung Semeru kind was used as the substrate; it was purchased from the Lumajang Regency's banana chip company. Despite the banana fruit is already mature, the banana skin is still green. The process of turning banana peel into flour involves numerous steps, including washing, slicing (to reduce size), drying, refining, and sifting. The banana peel was cleaned with distilled water before being sliced into approximately 2 cm × 2 cm pieces. The next step is to dry it out in the sun for the following three until six days. The samples of dried banana peel were then combined with a mixer and passed through a sieve with an 80 mesh size. The banana peel flour that was about to be used was first examined for its glucose content by Nelson Somogyi (Nelson, 1944), protein content using the Lowry method (Lowry et al., 1951), fat content using the Soxhlet method (AOAC 2005), moisture content using the gravimetric method, ash content using the sulfate method, nitrogen content using the alkalimetry, vitamin B1 content using the spectrophotometric method, and phosphate content using the spectrophotometric method.

Microorganisms and culture media

The main producer of L-lactic acid, *Lactobacillus plantarum* strain FNCC 0020, is based at the Center for Food and Nutrition Studies at Gajah Mada University in Yogyakarta. These pure cultures were inoculated and revived for growth on solid medium and slanted agar. The medium was made of MRS Agar (Himedia), which was heated to homogeneity using a hot plate and stirrer after being dissolved in distilled water in an amount of up to 2.5 grams. After that, the mixture was autoclaved for 15 minutes at 121° C to sterilize it. It was then moved to a test tube for liquid media and a petridish for solid media, respectively. This pure culture gets scratched onto each of these media after becoming firmly established. then kept at 37°C for 24 to 48 hours in an incubator. On MRS Broth liquid media, bacteria were grown, and the cultures were then shaken at 37 °C for 24 hours to create pure cultures.

Process the acid hydrolysis of Agung Semeru banana peel

The procedure that is currently being employed is the result of a modification where the primary step is the production of a hydrolyzate substrate. Banana Peel Flour was first sterilized in an autoclave for 15 minutes at 121 °C. Each variable mass also received 200 ml of sterile, distilled water with the following substrate concentrations: 2.5%, 7.5%, 12.5%, and 17.5%. After being heated at 80°C for 30 minutes, the product was filtered through a tofu or cheese filter to separate the filtrate. The filtrate was acid hydrolyzed by autoclaving at 121°C with a solution of 1% HCl v/v for 30 minutes (Pumiput et al., 2008). After hydrolysis, the hydrolyzate's pH was adjusted with $CaCO_3$ to 5–6 and the precipitate that had formed was separated using filter paper.

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Fermentation Lactic Acid

The pH-adjusted hydrolyzed filtrate was autoclaved for 15 minutes to sterilize it. Then, the 1% v/v culture of *the Lactobacillus plantarum* strain FNCC 0020 was added, and the mixture was incubated for 52 hours at 37°C and 100 rpm in a shaker incubator to begin the fermentation process. Following HPLC analysis, the best fermentation findings were chosen for the following variable, which involved varying the inoculum concentration employed (0.5% v/v, 1% v/v, 2% v/v, 4% v/v, and 5% v/v). Shimadzu HPLC, a UV-Vis Detector, and a Refractive Index were used to analyze the lactic acid concentration under the same process conditions as before.

RESULTS AND DISCUSSION

Semeru Agung peel flour characteristics

A chemical analysis technique called "proximate analysis" tries to determine the nutritional composition of a substance that is quantitatively assessed. The benefits of this analysis include the fact that it offers broad analytical results, does not require specialized technology for testing, may determine the total digestible nutrient (TDN) value, and can give an overall evaluation of how well a food ingredient is being used. Additionally, it has several flaws including its inability to appropriately calculate chemical composition levels and describe the texture and digestibility of food ingredients (Suparjo, 2010). Table 1 shows the findings of the proximate analysis of changes in banana peel flour.

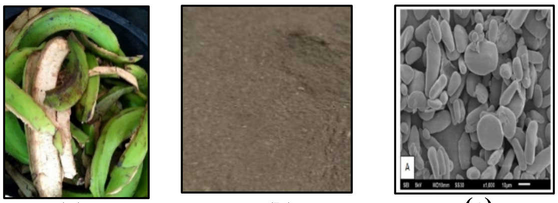
According to earlier research, the parameter values for each kind of banana peel flour provide distinct outcomes. Numerous variables, such as the type of analysis chosen, the morphology of the plants, the ripeness of the fruits, etc., have an impact on this. Depending on the type of process used utilizing certain tools, the proximate analysis of foodstuffs on each of these criteria varies. This is in line with a study that looked at how drying circumstances affected the physicochemical and antioxidant qualities of banana peels (Musa Cavendish), comparing sun and freeze-drying methods to drying ways employing appliances such a microwave, vacuum, dehumidifier, and hot air oven (Vu et al., 2017). Different techniques can be used to test carbohydrates, including the determination of total sugar or directly specialized techniques like starch, amylose, pectin, and lactose analysis. Analyzing fat similarly can be done using the Soxhlet, Babcock, acid hydrolysis, etc. Not much is different from other factors, such as protein content, water content, ash content, and fiber, which may all be analyzed using different types of methodologies and have an impact on the outcomes. Other variables include plant genetics, the soil in which they grow, climate, the physiological state of the fruit, and the methods used during harvest (packing, storage, and processing) (Morris et al., 2004). The morphology, agronomy, physiology, adaptability of accession to biotic and abiotic settings, etc., of the Agung Semeru (*Musa paradisiaca* L.AAB) banana plant have all been studied in research articles (Prahardini et al., 2016). When the growth reaches 80% of its growth period, changes in the fruit's content will occur, and they will thereafter diminish (Supriyadi, 2008).

Temperature, time, and the physiological maturity of agricultural products such as the time of harvest, the degree of ripeness, and others also affect value differences. The amount of water in the flour generated increases with the number of ripe bananas utilized. That indicated that as the fruit ripens, water content rises as a result of starch to sugar conversion. Additionally, the temperature and drying time has an impact on the water content that is created (Harefa & Pato, 2017). The proportion of fruit flesh to peels produced by this fruit depends on its degree of maturity. The increased level of ripeness creates a higher ratio of pulp to peels, which raises the moisture content of the pulp. This happens as a result of the respiration process, which converts starch into sugar and water. Due to the rising concentration of mineral salts, the level of fruit maturity also directly correlates with the ash content.

Table 1. Variations of banana peel flour based on proximate analysis

Nutrient parameter (%)	Banana peel flour variants								
	Kepok[a]	Kepok[b]	Kepok[c]	Raja[d]	Raja[e]	Tanduk[f]	Klutuk[g]	Uli[h]	Agung Semeru
Carbohydrate	72.88	62.91	62.65	74.1	87.9	79.9	86.5		70.52
Protein	8.33	7.26	7.08		2.9	2.9	8.2	6.76	5.68
Fat	14.85	12.23	12.51	5.32	1.2	1.1	2.7	1.18	3.115
Water	1.96	8.82	8.91	2.83				9.02	6.74
Ash	1.11	8.78	8.85	6.92					2.395
Fiber	2.83	51.21	51.93	1.81					13.38

Source: [a] (Hernawati, A. Aryani, 2017); [b] (Septiani & Srimiati, 2021); [c] (Anwar et al., 2021); [d] (Aryani et al., 2020); [e,f,g] (Sani, H. M., Ika, F. and Arta, 2015); [h] (Zahera, 2012)



(a) **(b)** **(c)**

Figure 1. (a) Agung Semeru Banana Peel; (b) Agung Semeru Banana Peel Flour; (c) SEM of Agung Semeru Banana Peel Flour (Hadisoewignyo et al., 2017)

As well undoubtedly with banana peels of Agung Semeru (*Musa paradisiaca* L.AAB) shows Figure1(a), the concentration of various mineral content in one type of banana fruit at various maturity levels (stage levels) has variable findings (Chandra et al., 2020; Ibiyinka et al., 2021; Khawas & Deka, 2016). This particular type of banana peel flour variant has an average parameter value of carbohydrate of 68.618%. The type of plantain peel flour, with a content of 74.13%, has the highest result for the carbohydrate parameter. In this investigation, the type of Agung Semeru banana peel flour had a carbohydrate content of 70.52 % in Table 1, which was fairly high. Based on sensory analysis, the Agung Semeru banana peel flour and starch's qualities comply with the needed standards for form, flavor, and odor. In contrast, microscopic analysis revealed that the banana peel starch generated also complied with the requirements, as shown by the existence of hyllus and lamellae. Banana starch has an eccentric or a point on a thin edge for its hyllus shape. The natural, minute particles known as granules are what gave starch its first form. There have been reports of single or independent grains having an elongated and cylindrical shape in the big banana peel starch. The large banana peel starch powder has particles in Figure 1 (c) that were 4.28 ± 0.22 m in size (dg). Compared to starch with a big grain size, smaller grain size starch will be less resistant to heating and water exposure (Hadisoewignyo et al., 2017). Other factors were also taken into account, including the following percentages of protein, fat, water, ash, and fiber: 5.68%, 3.115%, 6.74%, and 13.38% respectively. Banana peel flour's quality and durability depend on how much moisture it contains. Given that higher water contents make bacteria more likely to proliferate, the study's water content was within the acceptable range of 20% (Ministry of Health of the Republic of Indonesia, 1995). Among all the metrics, the carbohydrate parameter has the greatest value. In this study, high carbohydrate content is the factor with the greatest potential to influence the production of lactic acid during fermentation.

Figure 1 (b) depicts banana peel flour with a dark color that results from oxidation with air and the action of the food's enzymes (browning enzymatic). The oxidation reaction carried out by the enzyme polyphenol oxidase, commonly referred to as phenol oxidase, produces brown flour. These enzymes can catalyze the conversion of phenolic chemicals into quinones, which then polymerize to form the brown pigment melanin (Winarno, 2004). The starch or starch, a form of polysaccharide with the reaction depicted in Figure 1, is a type of carbohydrate found in banana peels. The properties of this starch are that it resembles a white powder, is insoluble in water, and is flavorless and odorless (Maitimu et al., 2020). Amylose and amylopectin are the two main monosaccharides in this polysaccharide type of starch. Maltose is created during the hydrolysis of starch molecules, while D-glucose is created when maltose is hydrolyzed (Winarno, 2004). The sweetness of the fruit reveals the amount of glucose from the starch that was created in it. Due to the climacteric nature of bananas, the ripening period or complex physiological changes (reversal of complex materials) may occur (Harefa & Pato, 2017). The ripeness stage of the fruit has an impact on its carbohydrate content as well.

Because more starch will convert to sugar as the fruit ripens, the sweetness of the fruit will increase with ripeness. Bananas are climacteric fruits, thus complex physiological changes or the ripening stage affect them as well. During the process, complex chemicals, including starch, are organized in cells. During this stage of ripening, the green hue will turn yellow, and the texture will soften, indicating a rise in sugar content but a decrease in starch (Sudjatha & Wisaniyasa, 2017).

Banana peel has the potential to be used as a carbon source in the lactic acid fermentation process by *Lactobacillus plantarum*, according to the results of the analysis of the features of banana peel flour. For lactic acid bacteria to use it to make lactic acid, high carbohydrate is a carbon source macronutrient for microbial growth (Abdel-Rahman et al., 2013). As an alternate source of starch, Agung Semeru banana peel flour can be utilized. The characterization of Agung banana peel flour revealed that it was consistent with the properties of starch in general with an excess of higher viscosity when compared to other starches, including cassava, corn, potato, tapioca, and wheat flour. In addition, the gelatinization temperature of the starch from the Agung banana peel was lower than that of cassava starch (Hadisoewignyo et al., 2017). For the growth of *Lactobacillus plantarum* strain FNCC 0020 in fermentation media, the starch content of this Agung banana peel flour serves as a carbon and energy source. Through the use of HCl in the hydrolysis process, starch will be broken down into simple sugars, which will then be fermented anaerobically in a closed system to produce lactic acid.

Effect of Substrate Concentration

Produced Agung Semeu banana peel flour was employed as a growth medium substrate for *Lactobacillus plantarum* strain FNCC 0020 in lactic acid fermentation. As the substrate concentration rises, lactic acid production rises, as seen in Figure 2. At substrate concentrations of 2.5% w/v, 7.5% w/v, 12.5% w/v, and 17.5% w/v, lactic acid was produced at levels of 1.885 g/L, 2.899 g/L, 4.925 g/L, and 5.401 g/L, respectively, while biomass production ranged from 0.564 g/L to 0.762 g/L to 1.520 g/L. The maximum level of lactic acid was discovered at a substrate concentration of 17.5%. This demonstrates that the amount of glucose that bacteria convert into lactic acid will grow in proportion to the number of substrates used or the concentration (Mladenović et al., 2019).

Lactobacillus plantarum growth could be considerably accelerated by adding rind flour, which contains 4.67% starch, to the growing media (Mohapatra et al., 2010). The presence of this substrate source will cause bacterial cells to expand, increasing the biomass as a result. The yield of lactic acid produced and its impact on substrate concentration have a significant impact on the number of bacterial colonies in the media. This increased substrate concentration of 75 mg/L produced the highest bacterial colony, 3E-09 CFU/ml, for 21 days at its peak condition until the stationary phase (Ferdaus et al., 2008). But the substrate also needs to have the right composition when it's being used as a source of carbs.

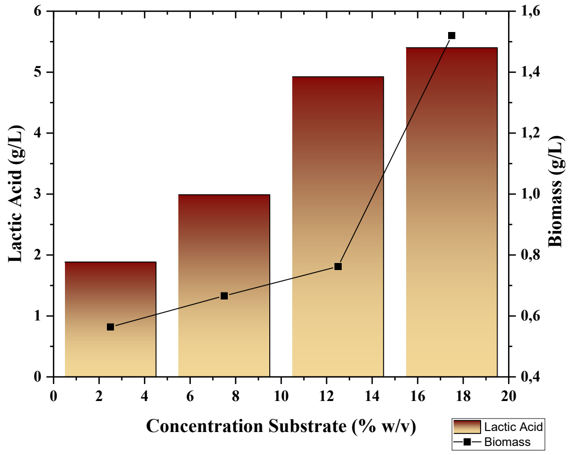


Figure 2. Effect of substrate concentration on lactic acid produced by *Lactobacillus Plantarum* strain FNCC 0020

The rate of lactic acid formation was substantially higher at the feed concentration of 65 g/L than it was at the substrate feeding rates of 95 and 115 g/L, indicating that the medium's glucose was not being fully utilized. This demonstrates the difficulty bacteria have in breaking down the sugar in pineapple juice, which slows down the pace at which bacteria multiply in the medium (Mochamad Busairi, 2008).

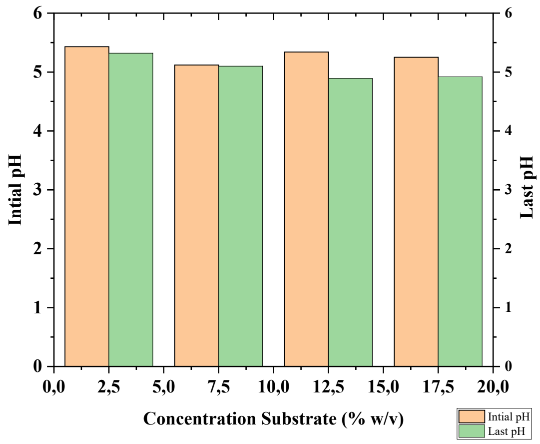


Figure 3. Effect of initial pH on final pH of lactic acid produced by *Lactobacillus Plantarum* strain FNCC 0020 at different substrate concentrations

A crucial parameter that has a significant impact on bacteria growth in the media, together with the substrate parameter, is pH. A reduction in pH and a rise in acidity will occur during the lactic acid production process. This is consistent with the bacterial activity depicted graphically in Figure 3, which is what generated it. The optimal pH range for the growth of *Lactobacillus plantarum* FNCC 0020 bacterium is 4.5–7.5, hence the medium needs to be adequate for this range to optimize the amount of product that is produced.

The addition of calcium carbonate ($CaCO_3$) can assist control the pH during fermentation so that the fermentation outcomes, specifically lactic acid, can be maximized. It is adequate to control the pH with different substrate modifications from Figure 3 in this fermentation process. Because it can maintain the fermentation process's level of acidity, calcium carbonate is a reagent that is frequently employed to neutralize lactic acid during fermentation. Determining the right quantities of $CaCO_3$ is crucial because 0.4% w/v levels will reduce the generation of lactic acid while 0.2% w/v levels have an increasing influence on lactic acid levels relative to no additions (Ferdaus et al., 2008).

Effect of Inoculum Concentration

Inoculum greatly influences the lactic acid produced, in Figure 4 shows the higher the inoculum concentration, the lower the lactic acid produced. At concentrations of 0.5% v/v, 1% v/v, 2% v/v and 4% v/v, the lactic acid was 8.586 g/L; 6.708 g/L; 4.693 g/L; and 2.680 g/L while the biomass is 1.032 g/L; 2.168 g/L; 2.564 g/L and 2.94 g/L. It also follows (Abdullah & Winaningsih, 2020). The highest lactic acid was obtained at an inoculum concentration of 0.5% v/v. Low concentration inoculum is added, which slows down fermentation but results in larger lactic acid production since the cells slowly convert sugar to lactic acid after they multiply. High inoculum concentration reduced cell viability and inhibited lactic acid generation (Asgher et al., 2010). This is also consistent with the fermentation of pineapple waste using *Lactobacillus delbruki*, where a low inoculum concentration of 5% yields the maximum lactic acid concentration of 54.97 g/L, compared to 10% and 15%, and decreases in production of lactic acid by 51.8 g/L and 44.84 g/L, respectively, over 168 hours compared to others (Abdullah & Winaningsih, 2020).

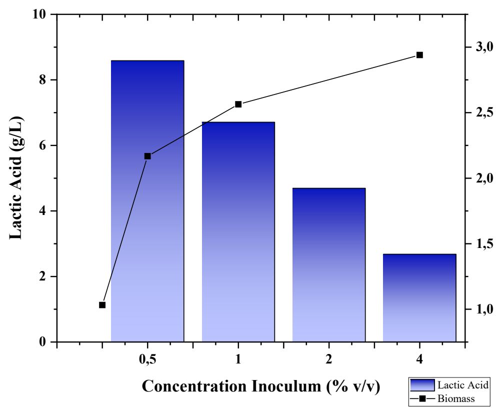


Figure 4. Effect of inoculum concentration on lactic acid produced by *Lactobacillus Plantarum* strain FNCC 0020

The addition of $CaCO_3$ is crucial because it can assist control the pH during fermentation so that more lactic acid is produced. This fermentation process with variations in inoculum concentration also can control the pH, as shown in Figure 5. The fermentation process by the bacterium Lactobacillus plantarum results in the production of the enzyme lactate dehydrogenase, which is greatly impacted by the ideal conditions for its maximum activity, one of which is pH. The pH level in the media will alter the activity of the enzymes, which will impact both the stability and speed of enzymatic activities (Ferdaus et al., 2008).

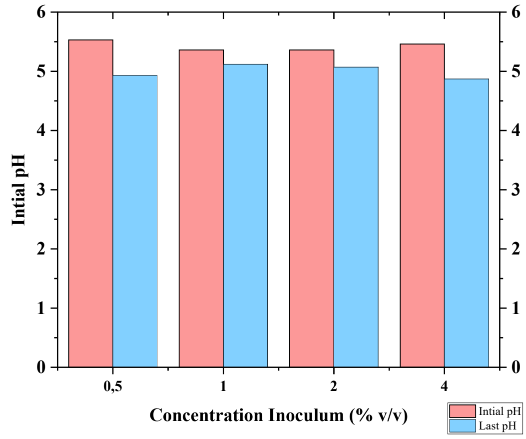


Figure 5. Effect of initial pH on final pH of lactic acid produced by *Lactobacillus Plantarum* strain FNCC 0020 at different inoculum concentrations

Effect Nitrogen Source

For the best possible outcome, lactic acid bacteria need a substrate with high nitrogen content. The most frequent supply of nitrogen for lactic acid fermentation is yeast extract because it gives the lactic acid bacteria an easy way to get the growth components they need. The concentration of supplements, particularly yeast extract, will rise concurrently with the creation of lactic acid. With the addition of 10 g/L of yeast extract, the maximum production rate was discovered from 2.31 g/L became 21.76 g/L (Abdullah et al., 2021). However, adding yeast extract to a large-scale fermentation process is not practical since the additional expenses for the fermentation process are more than the comparatively little amounts of lactic acid that are produced.

The nitrogen, sulfate, phosphate, and vitamin B1 concentrations of the Agung Semeru banana peel flour were analyzed to determine its original composition. Banana peel flour includes up to 0.8295% nitrogen, 0.037 gram sulfate, 1.6105% phosphate, and 0.2315% vitamin B1 in 100 grams. It is important to add the flesh flour to the bacterial culture medium because the amount of material currently presents is quite minimal. Other nitrogen sources, chosen based on quality and economic considerations, are utilized in place of yeast extract when nitrogen sources need to be replaced. Himedia RM027, a yeast extract, was employed as a standard for fair and accurate comparisons. According the product has a nitrogen content of 10.5% and a concentration of 10 grams/L, which conforms with the amounts of yeast extract that bacteria need. The concentration in the medium is 1.1 grams/L. Urea, which has a nitrogen content of 46.62 %, is equivalent to 2.395 g/L in the same medium, while ammonium sulfate (AS), which has a nitrogen content of 21.19%, is equivalent to 5.119 g/L, ammonium dihydrogen phosphate (ADP), which has a concentration of 13.13 %, is equivalent to 9.031 g/L, and a mixture of AS: ADP (2:1) is equivalent to 2.052 g/L.

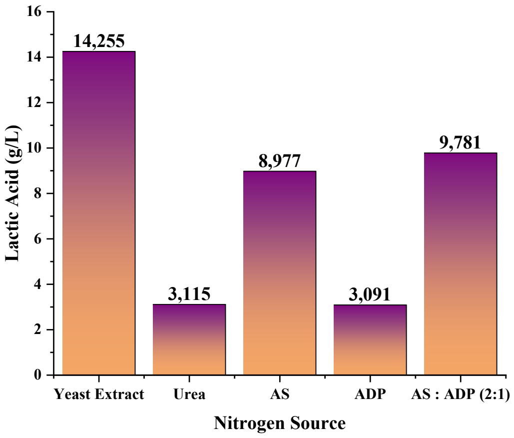


Figure 6. The effect of nitrogen type on lactic acid produced by Lactobacillus Plantarum strain FNCC 0020

The outcomes demonstrated that the inclusion of nitrogen sources has a beneficial impact to boost the production of lactic acid generated. Figure 6 shows the impact of several nitrogen sources with the same concentration ratio on the production of lactic acid. Yeast extract produced the most lactic acid during 52 hours, followed by AS: ADP, AS, ADP, and urea with yields of 14.255 g/L, 9.781 g/L, 8.977 g/L, 3.115 g/L, and 3.091 g/L, respectively. *Lactobacillus plantarum* FNCC 0020 is a bacterial yeast extract that is better suited to growing with additions in the media due to its maximal lactic acid production, as shown in Figure 6. Yeast extract contains a variety of other substances, including vitamins, sodium chloride, organic acids, peptides, amino acids, and dietary supplements. The mixture of ammonium sulfate and ammonium phosphate at a ratio of 2:1 produces the closest result to other nitrogen sources, and *Lactobacillus plantarum* FNCC 0020 bacteria tend to prefer it for growth. This further demonstrates that similar to the use of yeast extract, a rise in the concentration ratio will result in the production of lactic acid.

CONCLUSION

Lactobacillus plantarum FNCC 0020 bacterium may manufacture lactic acid from Agung Semeru banana peel flour as a source of carbohydrate substrate. The lactic acid produced from the substrate will be changed by the *Lactobacillus plantarum* FNCC 0020 bacterium into an increase in bacterial cell count. Lactic acid production will be decreased by an excessive amount of bacterial inoculum. The substrate and inoculum concentrations must be matched because they affect how quickly fermentation occurs and because ongoing cell development is influenced by the characteristics of the fermentation medium.

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