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Predicting Baker's Yeast Dosage and Fermentation Time on Bioethanol Production Made from Papaya Skin Waste

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ABSTRACT

This literature study research aims to find the appropriate yeast dose and fermentation time to process papaya skin waste into bioethanol by utilizing *Saccharomyces cerevisiae* yeast. Papaya skin waste is one of the biomass that is quite abundant and contains high sugar content so it can be used as raw material for making bioethanol. The data collection method was carried out through online searches of scientific journals related to the potential of papaya waste, fermentation, bioethanol production, bioethanol manufacturing techniques, yeast types and yeast doses, and bioethanol content. As a result, get fermentation for making bioethanol efficiently using a yeast dose of 0.3% and the fastest fermentation time of 3 days

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INTRODUCTION

Papaya (*Carica papaya L.*) is a semi-woody plant, usually single-stemmed, widely distributed in the tropics and subtropics (Jiménez et al., 2014). Current commercial varieties vary greatly in taste, possibly due to historically prioritized selection. for fruit appearance as well as great environmental effects (Zhou et al., 2021). Papaya contains essential nutrients and minerals which are the backbone of human survival (A. Rahman, 2013). Papaya is a tropical fruit that has commercial importance because of its nutritional and medicinal value (Saba & Pattan, 2016). The main challenge for the papaya industry is the inconsistency of fruit quality and, in particular, taste, which is a complex trait consisting of the perception of mouth taste (sweet, sour, or bitter) and the aroma produced by several volatile compounds (Zhou et al., 2021). The phytochemistry of C. papaya is increasingly being investigated and has shown great pharmacological promise as a result of its antioxidant, anti-inflammatory, immunomodulatory, and antimicrobial properties (Awais et al., 2021; Nafiu et al., 2018). Papaya fruit is a goldmine of vitamins C, E, K, minerals, and fiber and should ideally be consumed at least 4-5 servings a day (A. Rahman, 2013).

Papaya is much-loved by the public and is generally only used as a food ingredient in a fresh condition. Papaya fruit production tends to be dynamic where sometimes the abundant amount of fruit cannot be directly marketed (Simarmata et al., 2022). This fruit is used in the food industry for the production of jams, jellies, etc. As a result, the industry produces large quantities of papaya peels and papaya seeds as by-products, which are usually considered waste, and then disposed of. Papaya skin is a valuable source of bioactive compounds, which can be converted into many value-added products (Pathak et al., 2019). Papaya seeds and skin which are often discarded contain important nutrients that are beneficial to both humans and animals. The carbohydrate content of seeds and skins was 19.71% and 37.33%, respectively. The selected minerals revealed 6.43 mg/100 g and 16.23 mg/100 g of Calcium, 720.83 mg/100 g and 504.33 mg/100 g of Potassium, 4.21 mg/100 g and 2.73 mg/100 g (Moses & Olanrewaju, 2018). The fruit peels can be used in cosmetics, wastewater treatment, as animal feed, and as a binder in ceramics (A. Sharma et al., 2020).

It is recorded that almost 17% of food is wasted worldwide, this food can be used to produce by-products such as biofuels (Azad & Yesmin, 2019). One example is the trading of fruit in the market which generates a lot of waste because fruit has a short shelf life, so it spoils easily and tends to be disposed of as waste (Favaretto et al., 2023). Papaya fruit waste, for example, can be used as a raw material for making bioethanol through fermentation and distillation processes. The problem is that the duration of fermentation and heat treatment are important factors that affect the quality of bioethanol products because they affect the performance of yeast in the fermentation process (Fransisca, 2016). Papaya is a source of sugar and is one of the fruit wastes that can be used to produce bioethanol (Aunjt et al., 2016). Ripe papaya fruit is an ingredient for ethanol production from locally available fruits using simple, inexpensive, and adaptive technology (Parameswari et al., 2015). Moreover, papaya waste naturally undergoes fermentation and has the potential to be overgrown by microorganisms such as yeast (Utama, Sidabutar, et al., 2019).

Bioethanol produced from renewable biomass is expected to become one of the renewable energies that dominate biofuels in the transportation sector in the future, especially in the next twenty years (Nikolić et al., 2016). Bioethanol has been used in various countries as an alternative to gasoline, mainly due to its better emission characteristics (Kularathne et al., 2020). The choice of suitable, abundant, and cheap raw materials is very important because the cost of raw materials is a major part of the production costs (Nikolić et al., 2016). Furthermore, the remaining materials from the bioethanol production process can be used as fertilizers which are environmentally friendly, non-toxic, and biodegradable (Kularathne et al., 2020). The increasing need for bioethanol means that production must also be continuously increased by using raw materials that are cheaper and environmentally friendly. Based on these characteristics fruit waste can be considered cheaper and environmentally friendly (Dhanaseeli & Balasubramanian, 2014).

Ethanol production from agricultural products in the form of fruits can be increased by using genetically engineered yeast strains that can convert a lot of sugar into ethanol, through a process (Azad & Yesmin, 2019). The level and volume of bioethanol produced are greatly influenced by the duration of fermentation (Fransisca, 2016). The duration of fermentation affects reducing sugars, substrate pH, microbial biomass, and the resulting ethanol content (Sulfahri et al., 2016). An increase in the duration of fermentation does not produce results following an increase in ethanol production (Akponah & Akpomie, 2011).

Yeast is the most common microorganism used in bioethanol production and plays an important function in fermenting sugar into ethanol (Azhar et al., 2017). *Saccharomyces cerevisiae* is the most widely used yeast for ethanol production at an industrial level although ethanol is produced by various yeasts, bacteria, and fungi (Tesfaw & Assefa, 2014). Various types of yeast strains have been used in fermentation for ethanol production including hybrid, recombinant, and wild-type yeasts. Yeast can directly ferment simple sugars into ethanol while other types of raw materials must be converted into fermentable sugars before they can be fermented into ethanol (Azhar et al., 2017). It becomes a challenge then to estimate the duration of fermentation and the dose of yeast that must be used to obtain the maximum percentage of bioethanol. The composition of the fermentation time and the dose of yeast are important so that they can be used as a guide in the production of bioethanol made from papaya skin with fast, inexpensive results and maximum production.

METHODOLOGY

Articles were prepared using an information review approach in the form of a literature study. A literature study or literature study can be a simple summary with an organizational pattern, by combining summaries from various studies which are then made into a synthesis (Ramdhani et al., 2014). A literature study has a fundamental function in exploring theories or ideas that support and strengthen statements, establish and provide clear boundaries, and define and clarify the main concepts that will be used in the empirical study section (Nakano & Muniz, 2018).

The data or theory collected in this study relates to the potential of papaya waste, fermentation, bioethanol production, bioethanol production techniques, types of yeast, and dosage of yeast and bioethanol content. Data and theory were obtained through online searches of scientific journals with a very wide range, published between 2008-2023. Scientific journals are accessed via the website address https://scholar.google.co.id/ using several keywords such as papaya waste, bioethanol, fermentation time, yeast, and yeast dosage. Determination of the combination of fermentation time and dosage of yeast in the manufacture of bioethanol is important in efforts to produce fast, cheap, and produce maximum bioethanol.

The collected data and theory are then grouped to carry out mapping to facilitate analysis related to determining the combination of fermentation time and yeast dosage needed in the production of bioethanol. The collected theories are then combined to harmonize to give rise to a research statement. The results of this literature study are expected to contribute and provide additional information for further research and as a basis for exploring problems related to bioethanol production. This study is also expected to be a reference in the processing of papaya waste and its utilization as bioethanol.

RESEARCH RESULT

It is estimated that around 1.3 billion tons of food produced in the world for human consumption is wasted each year. The foods with the highest wastage are fruits and vegetables including tubers, which account for around 40-50% of global consumption (Deshbandhu, 2019). Fruit waste is available in abundant quantities, processing efforts are a way to recover the added value of products from this waste. Fruit waste is rich in sugar and carbohydrates which can be recovered and used for health in the manufacture of bioethanol (Jahid et al., 2018).

Potential Utilization of Papaya Waste to Become Bioethanol

Papaya is one of the most economical and valuable fruits in tropical countries. Unfortunately, the papaya fruit processing industry produces high by-products (Joymak et al., 2021). One of the by-products is papaya peel, which is usually considered waste, and then discarded (Pathak et al., 2019). Papaya waste causes significant commercial and environmental losses, mainly due to economic losses and bad smells when decomposed (Rojas-Flores et al., 2021). Papaya peel is a valuable source of bioactive compounds, which can be converted into many value-added products through fermentation such as biofuels, adsorbents, dietary fiber, biomedicine, and biomaterials (Pathak et al., 2019). Papaya pulp, skin, leaves, and other by-products can also be used as animal feed (Saran & Choudhary, 2013). Processing and utilization of papaya waste and by-products will also provide new opportunities for viable businesses and provide economic benefits for the papaya industry (Muzaffar et al., 2022).

Papaya waste and its by-products are an important source of bioactive compounds, vitamins, minerals, fiber, enzymes, sugars, and organic acids which can be converted into many nutraceutical products, and have various potential roles in the food industry and other different fields (Muzaffar et al., 2022). Papaya peel is one of the agricultural wastes that has enormous potential to be used as a promising raw material for bioethanol (Abdulla et al., 2018). The utilization of papaya skin is one of the strategies for minimizing waste and negative impacts on the environment (Abbaszadeh et al., 2016). Papaya waste naturally undergoes fermentation and has the potential to be grown by microorganisms such as yeast (Utama, et al., 2019). Papaya fruit skin is green when immature and changes from dark yellow to reddish-orange when ripe (Sharma & Kaur, 2017). Papayas in the green ripe and fully ripe stages showed higher maximum sugar content (4.8 g/100 g at 28 °C on day 12, and 10.2 g/100 g at 22 °C on day 8th) at higher temperatures (Yao et al., 2014).

The high sugar content in papaya skin makes it served as a superior raw material for bioethanol production (Abdulla et al., 2018). Bioethanol has great advantages over conventional fuels. Has a higher octane rating and is safer to use. The air quality will improve with clean and proper combustion (Jahid et al., 2018). The sugar content in papaya skin can be easily converted into simple sugars by the invertase enzyme found in *S. cerevisiae* (Abdulla et al., 2018).

Proximate analysis showed that papaya has high levels of protein (11.67 ± 0.04%), crude fiber (32.51 ± 0.03%), carbohydrates (47.33 ± 0.08%), ash (5.98%) ± 0.03 %) 0.03 %) and fat (2.51 ± 0.13 %). Mineral analysis shows that papaya peel is especially rich in potassium and phosphorus (Martial-Didier et al., 2017). The papaya skin also contains 2.54% oil and an ash content of up to 11.03% (Moses & Olanrewaju, 2018). Antioxidants present in various parts of papaya have free radical scavenging properties (Sharma & Kaur, 2017).

Utilization of Baker's Yeast in Making Bioethanol

Bioethanol is the most widely used biofuel in the world because it significantly contributes to reducing crude oil consumption and efforts to reduce environmental pollution (Azhar et al., 2017). Bioethanol is a potential alternative to conventional gasoline, especially because it has similar and superior qualities that allow for a reduction in greenhouse gas emissions and increases fuel reserves (Edeh, 2021). Bioethanol is considered the most promising alternative fuel, because it can be produced from a variety of agricultural-based renewable materials, such as bagasse (Setyawati et al., 2016). Production with commercial purposes that are made from food ingredients has an impact on increasing production costs (Edeh, 2021).

Bioethanol is produced by fermentation using microorganisms, one of which is yeast (Faustine & Djamaan, 2021). Through the fermentation process, yeast converts biomass into bioethanol (Naito et al., 2019). Yeast is widely used for the production of bioethanol from sugar-rich biomass (Hawaz et al., 2022). Yeast plays an important role in the production of bioethanol as an agent for fermenting sugars to ethanol. The great diversity of yeast isolated from plants, fruit, or parts of it can be a potential source (Hermansyah et al., 2021). Yeast can directly ferment simple sugars into ethanol while other types of raw materials must be converted into fermentable sugars before they can be fermented into ethanol (Azhar et al., 2017).

Saccharomyces cerevisiae is the most commonly used method of conventional bioethanol production (Hermansyah et al., 2021). This yeast can be fermented with several substrates from agricultural waste so it is environmentally friendly (Faustine & Djamaan, 2021). A wide variety of substrates have been used for ethanol production by utilizing the yeast *Saccharomyces Cerevisiae*, such as lignocellulose, molasses, sugar cane sorghum extract, starch-based substrates, and other wastes (Bhadana & Chauhan, 2016a). *Saccharomyces cerevisiae* is a common microbe used in ethanol production because of its high ethanol productivity, high ethanol tolerance, and ability to ferment a wide variety of sugars. However, some challenges in yeast fermentation inhibit ethanol production such as high temperature, high ethanol concentration, and the ability to ferment pentose sugar (Mohd Azhar et al., 2017).

Saccharomyces cerevisiae is the most widely used yeast for ethanol production at an industrial level although ethanol is produced by various yeasts, bacteria, and fungi (Tesfaw & Assefa, 2014). *Saccharomyces cerevisiae* is superior to bacteria, other yeasts, and filamentous fungi in various physiological characteristics regarding ethanol production in industrial contexts (Lin et al., 2014). This type of yeast is popular in ethanol production because its wide pH tolerance makes it less susceptible to infection. The ability of yeast to catabolize six-carbon molecules is the cornerstone for the production of bioethanol without proceeding to the end product of oxidation, namely CO2 (Khandaker et al., 2018).

Saccharomyces cerevisiae is generally considered safe for human consumption which increases its beneficial utilization over other yeasts and microorganisms (Tesfaw & Assefa, 2014).

Saccharomyces cerevisiae is a well-established organism for bioethanol production. The problem is that during the fermentation process, yeast cells are subjected to various stress conditions which inhibit their efficacy for commercial bioethanol production (Akhtar et al., 2018). The bioethanol fermentation process using yeast has several inhibiting factors such as high substrate concentration, ethanol as a product, and nutrients (Bahlawan et al., 2022). Bioethanol production during fermentation also depends on temperature, sugar concentration, pH, fermentation time, agitation speed, and inoculum size (Mohd Azhar et al., 2017). Efforts to coculture *Saccharomyces cerevisiae* with other microbes are targeted at optimizing ethanol production, shortening fermentation time, and reducing process costs. Yeast cell immobilization has been considered as a potential alternative to increase ethanol productivity (Bhadana & Chauhan, 2016b; Tesfaw & Assefa, 2014).

The main obstacle to the main production of bioethanol is the removal of the strong lignin component which is very resistant to dissolution and the main inhibitor for the hydrolysis of cellulose and hemicellulose. The pretreatment method has a significant impact on the efficient ethanol production from biomass (Tayyab et al., 2018). Pre-treatment is one of the important steps required to release fermentable sugars for use in microbial fermentation processes (Phwan et al., 2019). The alkaline pretreatment strategy can be an option for reducing cellulose crystallinity, reducing the lignin content to 84.83%, and increasing the cellulose content to 74.29% (Hernawan et al., 2017). Utilization of the cellulolytic activity of cellulose-degrading bacteria is another alternative to help convert cellulose into smaller sugars that will be more easily fermented by yeast (Promon, 2015).

Effect of Yeast Dosage and Fermentation Time on Bioethanol Yield

Two important stages in the production of bioethanol are the hydrolysis stage and the fermentation stage. Hydrolysis is the step of breaking the starch polymer chain into simple sugar units. The next stage is fermentation using microorganisms, such as Saccharomyces cerevisiae to convert sugar into ethanol. Bioethanol production in several studies stated that the fermentation stage is strongly influenced by the time and dose of the fermentor used. Efforts to evaluate through a literature review need to be carried out to study the fermentation time and fermentor doses that are effective in bioethanol production (Table 1). The hope is to get an effective dose combination and fast fermentation time to produce optimum bioethanol so that it can be used for bioethanol production using papaya skin.

Table 1. Combination of Dosage and Fermentation Time in Bioethanol Production

Dose Saccharomyces cerevisiae	Time	Bioethanol Content	Material	Temperature	Sugar Content	Literature sources
5%	8 day	14,17%	Cassava	-	28,18%	Candra et al., 2019
0,8%	3 day	17,98%	Ambonese banana peel	-	-	Rahmanto et al., 2022
4%	72 hour	9,38%	Coconut sap	30°C	1,54%	Gunam et al., 2022
0,8%	4 day	14, 61 %	Bagasse	30°C	32 g/100g	Maryana et al., 2020
25%	7 day	3.81%	Lemna minor (Duckweed)	-	-	Khodijah & Ahmad, 2015
0,3%	7 day	36%	Cassava	-	-	Kurniawan et al., 2014
0,9%	7 day	49,8%	Cassava	-	-	Kurniawan et al., 2014

Based on the results of a review of several studies (Table 1) it shows that the use of 0.8% yeast dose is included in the efficient use of materials and the shortest fermentation time (3 days) with a yield of 17.98% bioethanol. The most efficient dose is 0.3%, with a fermentation time of 7 days in cassava fermentation which produces bioethanol with a content of 36%. This means that the fermentation process for the manufacture of bioethanol efficiently can use a yeast dose of 0.3% and the fastest fermentation time is 3 days. This condition illustrates that the dose of Saccharomyces cerevisiae and the duration of fermentation greatly affect the level of bioethanol produced and can be used as an alternative combination of dosage and fermentation time in the manufacture of bioethanol made from papaya. This is in line with the view of Tuaputty (2020) which states that the concentration of the yeast used in the fermentation affects the increase in the volume and concentration of the bioethanol produced. According to Ahmad et al. (2019), the higher the concentration of Saccharomyces cerevisiae, the greater the level of bioethanol produced. The level of bioethanol is also affected by the fermentation time, where the longer the fermentation time, the higher the bioethanol content. Meanwhile, Rizaldi et al. (2022) confirmed that the high and low ratio of the amount of ethanol produced is influenced by sugar consumption and yeast growth during the fermentation process.

Bioethanol production on the other hand is also strongly influenced by the characteristics of the raw materials or biomass used. Production of bioethanol (alcohol) using biomass containing starch or carbohydrates, is carried out through the process of converting carbohydrates into sugars. Through the next fermentation process, the sugar is converted into bioethanol. This condition is shown by several research results with the same dosage and fermentation time for different raw materials to produce different concentrations of bioethanol (Table 2). This condition is in accordance with the opinion of Azhar et al. (2017) which states that yeast can directly ferment simple sugars into ethanol, while other types of raw materials must be converted into fermentable sugars before they can be fermented into ethanol. The composition of lignocellulosic biomass affects the production of reducing sugars. In lignocellulosic substrates, cellulose is physically bound to hemicellulose and chemically to lignin. Lignin and hemicellulose can inhibit the access of cellulase enzymes to cellulose (Narindri et al., 2016).

Table 2. Effect of Temperature and Sugar Content on Bioethanol Production

Dose *Saccharomyces cerevisiae*	Time	Bioethanol Content	Material	Temperature	Sugar Content	Literature sources
0,1 g	72 hour	3,5%	Jackfruit seeds	30°C	-	Kerina et al., 2022
0,1 g	72 hour	1,5%	Durian fruit seeds	30°C	-	
0,1 g	72 hour	4%	Mango fruit seeds	30°C	-	
6 g	6 day	7,39%	Oil palm empty bunches		17,10%	Rahman, 2022
25 g	9 day	70 %	Pineapple skin	78,4°C	-	Yusmartini et al., 2020
10 g	7 day	55%	Waste tapioca flour	27-32°C	17%	Amalia et al., 2021
2 g	72 hour	96,11 %	Papaya skin	30°C	-	Mitiku & Hatsa, 2020
2 g	72 hour	95,05 %	Mango skin	30°C	-	

2 g	72 hour	95,49 %	Banana skin	30°C	-	
4 g	96 hour	7%	Fiber cake	30°C	131 gr/L	Ahmad et al., 2019
0,0720 g	144 hour	13,53%	Banana kepok	28° C	3,170%	Febriana et al., 2018
0,0720 g	144 hour	12,99%	Banana raja	28° C	3,233%	
0,0720 g	144 hour	13,64%	Banana batu	28° C	3,219%	
0,4 g	5 day	2,51%	cherry fruit	-	-	Mukti & Aryani, 2016
10 g	72 hour	4.73%	Apple waste	28°C	-	Dhanaseeli & Balasubramanian, 2014
10 g	72 hour	6.21%	Grapes waste	30°C	-	
10 g	72 hour	4.19%	Papaya waste	27°C	-	
10 g	72 hour	5.4%	Banana waste	30°C	-	

There is also the influence of temperature and pH factors which affect the volume and concentration of the bioethanol produced. According to Azhar et al (2017), the production of bioethanol during fermentation depends on several factors such as temperature, sugar concentration, pH, fermentation time, agitation speed, and inoculum size. Temperature and pH affect the growth and activity of organisms in carrying out fermentation so that they have an impact on ethanol production (Ogbonda & Kiin-Kabari, 2013). The fermentation stage has an important role in increasing glucose and bioethanol levels linearly (Arif et al., 2018).

The yield of ethanol from fermentation depends on many factors. Fermentation efficiency and bioethanol yield depend on the raw materials, cultivars, and organisms used (Tse et al., 2021). Optimization of the most significant parameters such as pH, temperature, and fermentation time can be carried out to optimize bioethanol production and concentration (Tenkolu et al., 2022). The problem is that it is difficult to establish a general strategy that works for all types of biofuels originating from various metabolic pathways (Kang & Lee, 2015). One alternative strategy that can be implemented is S. cerevisiae coculture with other yeasts or target microbes to optimize ethanol production, shorten fermentation time, and reduce process costs (Tesfaw & Assefa, 2014).

CONCLUSIONS AND RECOMMENDATIONS

The sugar content in papaya skin makes it a potent ingredient to produce optimal volume and concentration of bioethanol. Based on the results of a literature study from various studies, it was found that fermentation for the manufacture of bioethanol efficiently can use a dose of 0.3% of the yeast Saccharomyces cerevisiae and the fastest fermentation time is 3 days. The combination of dosage and fermentation time can be an appropriate combination for the fermentation of papaya peel waste. Increasing the dose and fermentation time can be a strategy to increase bioethanol production. The challenge is that bioethanol production is also heavily influenced by the sugar content in the ingredients, temperature, and pH.

The results of this literature review can serve as a guideline for research and production of papaya peel-based bioethanol. Efforts to utilize papaya skin waste into bioethanol will be one of the strategies in utilizing papaya skin waste, thereby reducing waste accumulation and the impact of pollution that can be caused. Processing papaya skin waste into bioethanol is also a way to build a clean industry or implement the zero waste concept.

ADVANCED RESEARCH

This article is a literature review, so it needs to be followed up by conducting experiments or laboratory trials. In the future, experiments or laboratory experiments will be carried out not only based on the dosage and fermentation time, but also considering the sugar content in the ingredients, temperature, and pH.

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