

# **RESEARCH ARTICLE**

#### VALORIZATION OF CASSAVA PEELS WASTE THROUGH FERMENTATION PROCESS FOR **BIOETHANOL PRODUCTION: A REVIEW**

#### Abdullah and Nabilla Azzahra Eka Pramono

Department of Chemical Engineering, Diponegoro University, Semarang, Indonesia.

#### ..... Manuscript Info Abstract ..... Manuscript History Fermentation is one of the necessary processes in the production of Received: 10 October 2022 bioethanol. Reducing sugar from cellulose will convert it into Final Accepted: 14 November 2022 bioethanol with microorganisms as catalysts. Microorganisms used in Published: December 2022 bioethanol production also need to be considered and adjusted to the selection of the fermentation process. A previous study examined Keywords:bioethanol production. However, many still get relatively low vields Bioethanol, Cassava Peel, Fermentation, and require a long time of fermentation to produce bioethanol. Also Lignocellulose continuously developing to obtain bioethanol with high yield. The

fermentation process included Separate Hydrolysis Fermentation (SHF), Simultaneous Saccharification and Fermentation (SSF), and Consolidated BioProcessing (CBP). The fermentation process has its respective advantages and disadvantages, mainly when applied to lignocellulosic-containing materials. This review will explain and develop future trends in each method.

> Copy Right, IJAR, 2022, All rights reserved.

Introduction:-

Fermentation is a metabolic process that produces chemical changes in organic substrates through the action of enzymes. In biochemistry, it is narrowly defined as the extraction of energy from carbohydrates in the absence of oxygen. Food production may more broadly refer to any process in which the activity of microorganisms brings about a desirable change to a foodstuff or beverage [1]. Bioethanol is a form of renewable energy that can be produced from the fermentation of various feedstocks that contain sugars or carbohydrates, for example, rice, wheat, barley, potato, corn, and sugarcane. It is considered alternative fuel energy to replace fossil fuels. This firstgeneration bioethanol has gained attention, but its production competes with the food supply and land utilization. The subsequent generation has been made for producing bioethanol from nonedible feedstocks, including lignocellulosic biomass [2]. Using lignocellulosic biomass as raw material for bioethanol can reduce the scarcity or competition with food ingredients and provide its challenges. Lignocellulosic material comprises cellulose, hemicellulose, and lignin, one of the most abundant components in nature. It represents an extremely large quantity of renewable bioresources available on the planet having numerous applications [3]. Therefore, many studies have led to the utilization of energy from renewable energy sources, one of which is lignocellulosic material, as a raw material for the manufacture of bioethanol Figure.1.

Corresponding Author:- Nabilla Azzahra Eka Pramono Address:- Department of Chemical Engineering, Diponegoro University, Semarang, Indonesia.



Figure1:- Themanufactureofbioethanol.

Bioethanolis an effective sustainableenergysource.Bioethanolfuelscancontributetoacleanerenvironment and with the adoption of environmental protection laws in many countries, the demand for efficient bioethanol production processes may increase [4,5,6]. One of the important requirements is to have efficient microorganisms capable of fermenting various sugars as well as tolerating stressful conditions [7,8]. Bacterial and yeast strains that have ethanol-production properties have been developed through metabolic engineering. After several rounds of modification, three major microbial platforms, were Saccharomyces cerevisiae, Zymomonas mobilis, and Escherichia coli [9]. Indonesia is a fairly abundant producer of biomass, both from agricultural waste, plantation waste, and industrial waste such as empty palm oil bunches, corn cobs, sugar cane bagasse, sweet sorghum bagasse. and rice bran [10,11,12]. Accordingly, Indonesia has а greatopportunitytodeveloptechnologyforconvertingbiomassintoenergysources. This review will explain the characteristics of lignocellulose sources from cassava peel waste. Technology design and implementation from the conversion of simple sugars through the fermentation process to the multi-stage conversion of lignocellulosic material into bioethanol. The key to all research in this field is for reducing process costs thereby increasing the competitiveness of bioethanol against fuel oil earth (gasoline). The main factor being the cause is the high level of complexity that characterizes the processing of this material, so it requires pre-processing (pretreatment) to change the structure and chemical composition lignocellulose of tofacilitatehydrolysisefficiencycarbohydratesintofermentedsugars[13,14], as shown inFigure.2.



Fermentation methods are currently developing rapidly. The various types of lignocellulosic raw materials used result in various pretreatment methods. Various pretreatment strategies for producing bioethanol have alsobeenwidelyused. Various fermentation technologies for lignocellulosic materials have been tested and reviewed in this rticle.

#### CharacterizationofCassava Peel

Lignocellulosic materials consist mainly of three different polymers, known as lignin, hemicellulose, and cellulose, which are bonded together to form a unified whole. Each component's content depends on the type of biomass, age, and environmental conditions in which the biomass grows and develops. The characteristics oflignocellulosesources from cassavapeelwastewillbeexplainedinTable 1.

Table 1:- Characterizationolcassavapeei(Kel. [18]).					
ChemicalComponent(%)	CassavaPeels				
Ash Content	4.5				
HolocelluloseContent	66.0				
CelluloseContent	37.9				
HemicelluloseContent	37.0				
Lignin Content	7.5				

 Table 1:- Characterizationofcassavapeel(Ref. [18]).

Cellulose is the main component of lignocellulose in the form of microfibrils, a homopolysaccharide consisting of beta-D-glucopyranose linked to glycosidic bonds. In general, the structure of cellulose is crystalline, but there are also amorphous parts. So, the crystalline structure is very influential in the ability to hydrolyze chemically and also enzymatically [19]. Another carbohydrate source contained in lignocellulosic material is hemicellulose also known as polyose because it consists of various monomer sugars, namely pentose (close, rhamnose, and arabinose); hexoses (glucose, mannose, and galactose); and uronic acid (4-O-methyl glucuronic, D-glucuronic, and D-galacturonic). Hemicellulose has short polymer chains and is amorphous, so most of it is soluble in water.Therefore, hemicellulose isrelativelyeasytohydrolyze byacidstoform monomers[20].

#### Pretreatment

The pretreatment process is the initial treatment of materials before they are converted into product derivatives.Pretreatment aims to eliminate lignin that binds cellulose.The purpose of pretreatment is to open the structure of lignocellulosic cellulose to become more accessible to enzymes that break down polysaccharide polymers into sugar monomers. If not pretreated first, lignocellulose is difficult to be hydrolyzed because lignin is very strong in protecting cellulose so it is very difficult to hydrolyze before breaking down the lignin barrier.Obtained sugar without pretreatment is less than 20%, with pretreatment can increase to 90% of the yield theoretically [21].

Pretreatment	Description	Reference					
Biological	Biologicalpretreatmentprocessesincludefungalpretreatment, enzymatic	[22]					
	hydrolysis, and aeration. They are used to break down						
	thecrosslinkedstructuresoflignocellulosicwaste.						
Chemical	Thismethodisbasedontheuseofchemicalsforpretreatment, divided intofour	[23]					
	main methods:alkaline,acidic,ionic liquid, and organic solvent.						
Physical	Physical/mechanical pretreatment can open up the structure of LCBsby	[24]					
	disrupting their surface structure and reducing the size using						
	shearorcompressionforces.Physicalpretreatmentsincludemilling,sonication,						
	mechanical extrusion, ozonolysis, and pyrolysis.						
Physicochemical	Combining fungal pretreatment with other physical and chemical methods [2						
	isawaytoovercomethisdrawback.						

 Table 2: Varioustypesofpretreatmentsusedinlignocellulosic

#### Hydrolysis

Hydrolysis or saccharification is the process of breaking down polysaccharides in lignocellulosic biomass, namely cellulose and hemicellulose into their constituent sugar monomers. Complete hydrolysis of cellulose produces

glucose, while hemicellulose produces several monomers of sugars in pentose ( $C_5$ ) and hexose ( $C_6$ ). Hydrolysis can becarried out chemically (academically) or enzymatically [26].

#### Fermentation

Fermentation is the process of chemical changes in organic substrates, whether carbohydrates, proteins, fats, or others, through the activity of biochemical catalysts known as enzymes produced by specific microbes. In summary, their action for converting glucose to ethanol:

$$C_6H_{12}O_6 \rightarrow 2C_2H_5OH + 2CO_2$$
[27]

Specific microbes that are oftenusedinindustrial ethanolfermentationare *Saccharomyces cerevisiae*, *S.uvarum*, *Schizosaccharomyces sp.*, and *Kluyveromces sp. Saccharomyces cerevisiae* is a microbial species (yeast) anaerobic that has a very high conversion power of sugar into ethanol. This microbe is commonlyknown as baker's yeast and its metabolism has been well studied. The main products of metabolism are ethanol, carbon dioxide, and water, while some other products are produced in small amounts.Saccharomyces cerevisiaerequiresatemperatureof30°CandapHof4.0-5togrowwell[28].

# Methode:-

### Separated Hydrolysis Fermentation (SHF)

SHF is one of the configurations that have been tested more extensively. Pentose fermentation, when it is carried out, is accomplished in an independent unit. In the SHF configuration, the joint liquid flow from both hydrolysis reactors first enters the glucose fermentation reactor. The mixture is then distilled to remove the bioethanol leaving the unconverted xylose behind. In a second reactor, xylose is fermented to bioethanol, and the bioethanol is again distilled. The advantage of SHF is the ability to carry out each step under optimal conditions, i. e. enzymatic hydrolysis at 45-50 °C and fermentation at about 30°C but this approach proves to be very costly [29]. So based on the different combinations of technologies adopted at the pretreatment, hydrolysis, and fermentation stages of ethanol synthesis, several integrated technologies have been developed. In the below, existing integrated conversion technologies are discussed.

Ref	Raw Material	<b>Operating Conditions</b>	Results
[30]	Cassavaplantsvariet y Sree Jaya(such as stems,leaves,and peels)	Pretreatmentusingmicrowaveirradiationfor 7minutesassistedby20galkali pretreatment in200 ml3%NaOH.Fermentationusing <i>Saccharom</i> <i>yces</i> <i>cerevisiae</i> andthefermentationprocesswasca rriedoutfor168h(120hforsaccharification+4 8hoursforfermentation)atroomtemperature( 30±1°C).	The high est percentage of reducing sugaru tilization during fermentation (48h) was 91.02% in stems. The highest ethanol yield (ml/kg dry biomass) was 303 for the peel.
[31]	Cassava Pulp	FermentationusingSaccharomyces cerevisiae, Trichoderma reesei, Bacillus licheniformis, Aspergillusniger, andcocktail enzymes.Thetemperaturewassetto37°Can dpH5for120hours.	The ethanol produced is29.39g/L
[32]	Cassava Peel	The powdered starch of Cassava peelwas hydrolyzedduringPEFB-SO3H in a 250mLglass reactor.FermentationusingSaccharomyces cerevisiaeBY4743 with a temperature of 30°Cfor72hour,speed (50,100,150)rpm,andpH(4,4.5,5).	The best result was found at pH 4.5 and 50 rpm for a 24 h reaction with 3.75 g/Lofbioethanol concentration.
[34]	Cassava andsweet potatopeels	FermentationusingGloeophyllum sepiarium and Pleurotus ostreatusfor hydrolysis and Zymomonas mobilis and	50g of cassava peel and 50g of sweetpotatopeelyieldethanolof11.97g/c $m^3$ (26%)and6.5g/cm <sup>3</sup> (12%).

 Table 3: PreviousstudiesofmakingbioethanolusingtheSHFprocesswithcassavawasteasrawmaterials.

Saccharomyces cerevisiaeforfermentation. The temperatureusedwas 28°C with7 days
forhydrolysis and5daysforfermentation

### Simultaneous Saccharification and Fermentation (SSF)

Saccharification and fermentation are carried out simultaneously in a single reactor, thus allowing for cost-saving and reduction of inhibitors, increasing the hydrolysis rate. The critical issue of this solution is the optimization of process conditions concerning both enzymes and microorganisms at the same time. The key to the SSF process from biomass is its ability to rapidly convert the sugars into ethanol as soon as they are formeddiminishing their accumulation in the medium. Bearing in mind that the sugars are much more inhibitory for the conversion process than ethanol, SSF can reach higher rates, yields, and ethanol concentrations compared to the SHF process.SSF offers an easier operation and a lower equipment requirement than the sequential process since no hydrolysis reactors are needed; moreover, the presence of ethanol in the broth makes the reaction mixture less vulnerable to the action of undesired microorganisms. Nevertheless, SSF has the inconvenience that the optimal conditions for fermentation implies hvdrolvsis and are different. which а difficult control and optimization of process parameters; in addition, larger amounts of exogenous enzymes are required. Saccharification with cellulolytic enzymes is best done at around 50 °C, while most fermenting microbes have an optimum temperature for ethanolfermentation between 28 °C and 37 °C [35]. In practice, it would bedifficult to lower the optimum temperature of cellulases through proteinengineering. Accordingly, hightemperature fermentation is in high demand for simultaneous saccharification and fermentation, and thermotolerant yeast trainshavebeenscreenedfortheabilitytofermentethanol. *Kluyveromycesmarxianus* appears to be particularly promising. Many strains of K. marxianus grow well at temperatures as high as 45-52 °Candcanefficiently produce ethanol at temperatures between 38 °C and 45 °C.Moreover, K.marxianusoffers additionalbenefits including a high the rate and ability utilize а wide variety substrates growth to of sugar (e.g., arabinose, galactose, mannose, xylose) at elevated temperatures [36].

Ref.	RawMaterial	Operating Conditions	Results
[37]	Cassava	Fermentationusing Saccharomycescere	Bioetanol concentration
	peelwaste	<i>visiae</i> , termamyl (α-amylase)	16,42±0,26g/L.
		120L, amyloglucosidase (glucoamylase	_
		), and cellulite enzyme	
		1.5L.Operatedatatemperatureof33.73°	
		CandpH5.31.	
	Cassava	Bioethanolproductionfromalkali-	The ethanol produced was 0.44 g/g
[38]	peelwaste	assistedhydrothermalpretreatedcassava	with a fermentation efficiency of
		peelbythermotolerantKluyveromycesm	86.11%.Theresultingreducedsugarwas
		arxianusMTCC4139strain,combinedw	670.58±10.13mg/gwithasaccharificati
		ith( $\alpha$ -amylase, glucoamylase, and	on efficiency of 81.25% ±3.20%.
		cellulase).TheSSFwasperformed at	
		100 rpm,40°C for a 72h period.	
[39]	Cassava pulp	FermentationusingSaccharomycescere	The ethanol produced is43.35g/L.
		visiae, Trichodermareesei, Bacilluslich	
		eniformis, Aspergillusniger, and cocktail	
		enzymes.Thetemperaturewassetto37°C	
		andpH5 for120h.	
[40]	Cassava pulp	FermentationusingSaccharomycescere	Theyieldof0.38gethanol/gCP,andethan
		visiaeTISTR5339andKomagataeibact	olproductionrateof0.309 g/h.
		ernataicolaTISTR998.Enzymeuseda-	
		amylaseandglucoamylaseratio(75:25).	
		Thefermentationtemperaturewaskeptc	
		onstantat30°Cfor120h.	
[41]	Cassava pulp	Fermentation usingSaccharomyces	Theyieldwas27.4 g/L of ethanol.
		cerevisiae.Enzymeuseda-	

Table	4.	Dave			£ a 1 .:		1	1	41 C CT		1		
Table 4		Prev	/ioussii	laieso	Imaki	ngoi	bethand	Jusing	inessr	processwi	incassav	awasteasrav	vmaterials.

#### **Consolidated BioProcessing (CBP)**

All enzymes and bioethanol are produced in a single reactor by a single microorganism community. The logical culmination of reaction-reaction integration forthetransformationofbiomassintoethanolisconsolidatedbioprocessing (CBP), known also as direct microbial conversion (DMC). The key difference between CBP and the other strategies of biomass processing is that only one microbial community carries out both the production of cellulases and fermentation, i.e., cellulase production, cellulose hydrolysis, and fermentation are carried out in a single step. This difference has an important advantage as no capital or operation expenditures are required for enzyme production within the process. Similarly, part of the substrate has not deviated from the production of cellulases. Moreover, the enzymatic and fermentation systems are entirely compatible. Thermophilic cellulolytic anaerobic bacteria have also been extensively examined for their potential as bioethanol producers. These bacteria include Thermoanaerobactere thanolicus, Clostridium thermohydrosulfuricum, Thermoanaerobactermathranii, Thermoanaerobiumbrockii, Clostridiumthermosaccharolyticum strain, etc. including others [42]. Thermophilic cellulolytic anaerobic bacteria have a distinct advantage over conventional yeasts for bioethanol production in their ability to directly use a variety of inexpensive biomassfeedstocksandtheirabilityto withstand temperatureextremes.Thelowbioethanoltolerance of thermophilic anaerobic bacteria (<2%, v/v) is a major obstacle to their industrial exploitation for ethanol production [43]. Cell surface engineering has been applied to a thermotolerant strain of the yeast K.marxianus forthedisplayofcellulolyticenzymesonthecellsurface. Recombinant K. marxianus straincodisplaying endoglucanase and β-glucosidase on the cell surface grew well at temperatures as high as 48 °C, which ethanol was produced from the cellulosic material  $\beta$ -glucan with a yield of 0.47 g ethanol per gram of consumed carbohydrate [44]. This study gives supports the development of CBP yeast for effective bioethanol production. Another proposed approach is the utilization of mixed cultures in such a way that the hydrolysis and fermentation of lignocellulosic biomassbe carried out simultaneously.

Ref	Raw Material	Operating Conditions	Results
[45]	Cassava	Single-	Thisethanolwasproducedataconcentrati
	starchhydrolysis	stepfermentationbyS.cerevisiaeat34°C	on of 81.86 g/L (10.37%
	andfermentation	ina5-Lfermenter.	v/v)withayieldcoefficientof0.43
			g/g,productivityat 1.14 g/L/h,and an
			efficiency of 75.29%.
[46]	Cassava pulp	Engineered S. cerevisiae Kyokai strainK7	91% of the theoretical yield.
		(K7G). Recombinant: cell surface	
		engineering system, displaying oryzae	
		glucoamylases.	
[47]	Cassavastem	Fermentation using	Bioethanolproduced7.84±0.31g/Lbioet
		ClostridiumthermocellumATCC31,924.	hanolwith62.37±0.25% cellulose
		Pretreatmentusingdilutealkali.	conversionefficiency.
[48]	Cassava	Fermentation using	79.75 and 69.73
	starchandrawcassav	K.marxianusstrainYRL009.Reco	g/lfromcassavastarchandrawcassavapo
	atuberpowder	mbinantexpressing α-	wder, respectively.
		amylasefromA.oryzaeas well as αamylase	
		andglucoamylasefromD.occidentalis.	

 Table 5: PreviousstudiesofmakingbioethanolusingtheCBPwithcassavawasteasrawmaterials.

# **Conclusion and Future Trends:-**

The efficient utilization of lignocellulosic biomass is very important for the further development of bioethanol. In addition, using lignocellulosic biomass will help reduce people's dependence on non-renewable resources and reduce competition in using raw materials for food sources. In summary, many possibilities for using lignocellulosic biomass especially cassava peel waste have been proposed in recent years, and promising results have been achieved. The selection of the fermentation process method will affect the product's quality. Undesirable conditions during the fermentation process will result in the formation of other derivative products

and a separation process is required. From the method that has been conveyed, it is found that the use of the SSF method can produce ethanol in greater quantities and reduce the cost of the equipment. from the existing ethanol yield and good yield. some of the methods that have been described in this review are very helpful for researchers in selecting the fermentation method and needfurtherresearchforotheragricultural wastes.

# **References:-**

- 1. Hui, Y. H. Handbook of vegetable preservation and processing. New York: M. Dekker. (2004), p. 180.ISBN 978-0-8247-4301-7. OCLC52942889.
- Srinophakun, Penjit; Thanapimmetha, Anusith; Srinophakun, Thongchai Rohitatisha; Parakulsuksatid,Pramuk;Sakdaronnarong,Chularat;Vilaipan,Monsikan;Saisriyoot,Maythee.Techno-Economic Analysis for Bioethanol Plant with Multi Lignocellulosic Feedstocks. International Journal ofRenewableEnergyDevelopment, 9(3),319–328 (2020). doi:10.14710/ijred.9.3.319-328.
- 3. S.Kumar; S. P. Singh; I. M. Mishra; D. K. Adhikari. Recent Advances in Production ofBioethanolfromLignocellulosicBiomass.32(4), 517–526 (2009).doi:10.1002/ceat.200800442.
- 4. Mágda CorreiaSantos, Daniel Fernandes Costa, Allan Almeida Albuquerque, João InácioSoletti, Simoni Margareti Plentz Meneghetti, Alternative distillation configurations for bioethanol purification: Simulation, optimization, and techno-economic assessment, Chemical Engineering Research, and design, Volume 185, (2022), pages 130-145, ISSN0263-8762. https://doi.org/10.1016/j.cherd.2022.06.036.
- 5. ChunShengGoh;KokTatTan;KeatTeongLee;SubhashBhatia. Bio-ethanol from lignocellulose:Status,perspectives,and challenges in Malaysia.101(13),4834–4841 (2010).doi:10.1016/j.biortech.2009.08.080.
- 6. Khatiwada,D.AssessingthesustainabilityofbioethanolproductioninNepal(Licentiatedissertation,KTHRoyalI nstituteofTechnology) (2010).Retrievedfromhttp://urn.kb.se/resolve?urn=urn:nbn:se:kth:diva-25336.
- Upasana Jhariya; Nishant A. Dafale; Shweta Srivastava; Rahul S. Bhende; Atya Kapley; Hemant J.Purohit. Understanding Ethanol Tolerance Mechanism in Saccharomyces cerevisiae to EnhancetheBioethanolProduction:CurrentandFutureProspects.BioEnergyResearch,14:670-688 (2021).doi:10.1007/s12155-020-10228-2.
- Juhi Sharma, Vinod Kumar, Rajendra Prasad, Naseem A. Gaur. Engineering of Saccharomyces cerevisiae as a consolidated bioprocessing host to produce cellulosic ethanol: Recent advancements and current challenges,BiotechnologyAdvances,Volume56,107925,ISSN0734-9750 (2022).https://doi.org/10.1016/j.biotechady.2022.107925.
- 9. J. Zaldivar; J. Nielsen; L. Olsson. Fuel ethanol production from lignocellulose: a challenge formetabolicengineeringandprocess integration. 56(1-2),17–34 (2001).doi:10.1007/s002530100624.
- 10. Dani, S., & Wibawa, A. Challenges and policy for biomass energy in Indonesia. International Journal of Business, Economics, and Law, 15(5), 41-44 (2018).
- 11. Syaifuddin Yana, Muhammad Nizar, Irhamni, DewiMulyati, Biomass waste as renewable energy in developing bio-based economies in Indonesia: A review, Renewable, and Sustainable EnergyReviews, Volume160, 112268, ISSN1364-0321 (2022), https://doi.org/10.1016/j.rser.2022.112268.
- 12. Guild, James. Feed- in- tariffsandthepoliticsofrenewableenergyinIndonesiaandthePhilippines. Asia&thePaci ficPolicyStudies, 6(3), 417–431 (2019). doi:10.1002/app5.288.
- Ruiz, Héctor A.; Hedegaard Thomsen, Mette; Trajano, Heather L. Hydrothermal Processing inBiorefineries||HydrothermalPretreatmentof LignocellulosicBiomassforBioethanolProduction.10.1007/978-3-319-56457-9(Chapter7),181–205 (2017). doi:10.1007/978-3-319-56457-9 7.
- 14. Toor, Manju; Kumar, SmitaS.; Malyan, Sandeep K.; Bishnoi, Narsi R.; Mathimani, Thangavel; Rajendran, Karthik; Pugazhendhi, Arivalagan. An overview of bioethanol production from lignocellulosic feedstocks. Chemosphere, 125080 (2019). doi:10.1016/j.chemosphere.2019.125080.
- 15. Mustafa Balat. Production of bioethanol from lignocellulosic materials via the biochemical pathway: Areview. 52(2), 858-875 (2011). doi:10.1016/j.enconman.2010.08.013.
- 16. Nathan Mosier; Charles Wyman; Bruce Dale; Richard Elander; Y.Y. Lee; Mark Holtzapple; MichaelLadisch. Features of promising technologies for pretreatment of lignocellulosic biomass. 96(6),673–686 (2005).doi:10.1016/j.biortech.2004.06.025.
- 17. Hsu, T.A., Ladisch, M.R. and Tsao, G.T. Alcohol from Cellulose. Chemical Technology, 10,315-319 (1980).
- 18. Daud, Zawawi; Awang, Halizah; Kassim, Angzzas Sari Mohd; Hatta, Mohd Zainuri Mohd; Aripin, Ashuvila Mohd. Comparison of Pineapple Leaf and Cassava Peel by Chemical Properties and

morphology characterization. Advanced Materials Research,974,384–388 (2014). doi:10.4028/www.scientific.net/AMR.974.384.

- 19. JitendraKumarSaini;ReetuSaini;Lakshmi Tewari.Lignocellulosic agriculture wastes asbiomassfeedstocksforsecond-generation bioethanol production:conceptsandrecentdevelopments (2015).
- 20. Martina Andlar; Tonci Rezi; Nenad Mardetko; Daniel Kracher; Roland Ludwig; Bozidar Santek.Lignocellulose degradation: An overviewof fungiand fungal enzymes involvedinlignocellulosedegradation (2018).
- 21. Carlo N Hamelinck; Geertje vanHooijdonk; André PC Faaij. Ethanol from lignocellulosicbiomass:technoeconomicperformanceinshort-, middle-and long-term.28(4),384–410 (2005).Doi:10.1016/j.biombioe.2004.09.002.
- 22. Efeovbokhan, V. E., Egwari, L., Alagbe, E. E., Adeyemi, J. T., & Taiwo, O. S. Production of bioethanol from hybrid cassava pulp and peel using microbial and acid hydrolysis. BioResources, 14(2),2596-2609 (2019).
- Klinpratoom, Bunpot; Ontanee, Anissara; Ruangviriyachai, Chalerm. Improvement of cassavastemhydrolysisbytwo-stagechemicalpretreatmentforhighyieldcellulosicethanolproduction.KoreanJournalofChemicalEngineering,32(3),413–423 (2015).doi:10.1007/s11814-014-0235-8.
- 24. Pooja,N.S.;Padmaja,G.Enhancing the EnzymaticSaccharificationofAgriculturalandProcessing Residues of Cassava through Pretreatment Techniques. Waste and Biomass Valorization,6(3),303–315 (2015).doi:10.1007/s12649-015-9345-8.
- Kristiani, Anis; Effendi, Nurdin; Aristiawan, Yosi; Aulia, Fauzan; Sudiyani, Yanni. Effect of Combining Chemical and Irradiation Pretreatment Process to Characteristic of Oil Palm's Empty FruitBunches as Raw Material for Second Generation Bioethanol. Energy Procedia, 68, 195–204 (2015). doi:10.1016/j.egypro.2015.03.248.
- 26. S Arita; F Hadiah; R Amalia; E Rosmalisa; W Andalia. Production of Glucose from Waste BarkAcaciaMangiumUsingDelifnificationandChemicalHydrolysis Process (2019).
- 27. Medina, R. Fermentation Technology. Britania Raya: EDTECH (2019).
- 28. GraemeMWalker;GrahamGStewart,SaccharomycescerevisiaeintheProductionofFermentedBeverages (2016).
- 29. Charlotte Tengborg; Mats Galbe; Guido Zacchi. Influence of Enzyme Loading and PhysicalParametersontheEnzymaticHydrolysisofSteam-PretreatedSoftwood.17(1),110–117 (2001).doi:10.1021/bp000145+.
- Pooja, N. S.; Sajeev, M. S.; Jeeva, M. L.; Padmaja, G. Bioethanol production from microwaveassistedacidoralkalipretreatedagriculturalresiduesofcassavausingseparatehydrolysisandfermentation(SHF).3Biotech,8(1), 69 (2018).Doi:10.1007/s13205-018-1095-4.
- 31. Mingjun ZHU, Ping LI, Xinfang GONG & Jufang WANG. A Comparison of the Production of EthanolbetweenSimultaneousSaccharificationand Fermentation and SeparateHydrolysis and fermentation Using Unpretreated Cassava Pulp and Enzyme Cocktail, Bioscience, Biotechnology, and biochemistry, 76:4, 671-678 (2012), DOI:10.1271/bbb.110750.
- 32. BCChoo;KSKIsmail;AHMa'Radzi.Scaling-upandtechno-economicsofethanolproduction from cassava starch via separate hydrolysis and fermentation. IOP Conference Series: EarthandEnvironmentalScience(2021). doi:10.1088/1755-1315/765/1/012004.
- 33. Mardina, P., Irawan, C., Putra, M. D., Priscilla, S. B., Misnawati, M., & Nata, I. F. BioethanolProduction from Cassava Peel Treated with Sulfonated Carbon Catalyzed Hydrolysis. Jurnal KimiaSains danAplikasi,24(1), 1-8 (2021). https://doi.org/10.14710/jksa.24.1.1-8.
- Oyeleke, Solomon Bankole & Dauda, B.E.N. & Oyewole, Oluwafemi & Okoliegbe, I.N. & Ojebode, T. Production of bioethanol from cassava and sweet potato peels. Advances in EnvironmentalBiology.5.3729-3733 (2011).
- 35. Eryati Derman; Rahmath Abdulla; Hartinie Marbawi; Mohd Khalizan Sabullah; Jualang Azlan GansauPogaku Ravindra, Simultaneous Saccharification and Fermentation of Empty Fruit. Bunches ofPalmforBioethanolProductionUsingaMicrobialConsortiumofS.cerevisiaeandT.harzianum (2022).
- 36. DungMinhHa-Tran;TrinhThiMyNguyen;andChieh-ChenHuang.Kluyveromycesmarxianus:CurrentStateofOmicsStudies,StrainImprovementStrategy andPotentialIndustrialImplementation (2020).
- 37. Aruwajoye, Gabriel S.; Sewsynker-Sukai, Y.; Kana, E.B. Gueguim. Valorization of

cassavapeelsthroughsimultaneoussaccharificationandethanolproduction:Effectofprehydrolysistime,kinetica ssessment, andpreliminaryscaleup.Fuel,278,118351 (2020).DOI:10.1016/j.fuel.2020.118351.

- 38. Narendra Kumar Papathoti; Kansinee Laemchiab; Vineela Sai Megavath; PraveenKumar Keshav;Parichat Numparditsub; Toan Le Thanh; Natthiya Buensanteai. Augmented ethanol production from alkali-assisted hydrothermal pretreated cassava peel waste. Energy Sources, Part A: Recovery,Utilization, and environmental effects (2021). doi:10.1080/15567036.2021.1928338.
- 39. Mingjun ZHU, Ping LI, Xinfang GONG & Jufang WANG. A Comparison of the Production of EthanolbetweenSimultaneousSaccharificationandFermentationandSeparateHydrolysisandFermentation Using Unpretreated Cassava Pulp and Enzyme Cocktail, Bioscience, Biotechnology, and biochemistry, 76:4, 671-678 (2012), DOI:10.1271/bbb.110750.
- 40. SiriwanKhanpanuek,SiripornLunprom,AlissaraReungsang,ApilakSalakkam, Repeated-batch simultaneous saccharification and fermentation of cassava pulp for ethanol production using amylases and Saccharomyces cerevisiae immobilized on bacterial cellulose, Biochemical Engineering Journal,Volume177, (2022)108258,ISSN1369-703X,https://doi.org/10.1016/j.bej.2021.108258.
- 41. Siriwong, Tanyaporn; Laimeheriwa, Bustomi; Aini, UyunNurul; Cahyanto, MuhammadNur; Reungsang, Alissar a; Salakkam, Apilak. Coldhydrolysisofcassavapulpand its useinsimultaneous saccharification and fermentation(SSF) processforethanolfermentation. Journal ofBiotechnology, (2019), S0168165619300070. doi: 10.1016/j.jbiotec. 2019.01.003.
- 42. Mustafa Vohra; Jagdish Manwar; Rahul Manmode; Satish Padgilwar; Sanjay Patil, Bioethanolproduction:Feedstockandcurrenttechnologies (2013).
- 43. Jan Eric Jessen; Johann Orlygsson. Production of Ethanol from Sugars and LignocellulosicBiomass byThermoanaerobacterJ1IsolatedfromaHotSpringinIceland (2012).
- Mejía-Barajas, J. A., Alvarez-Navarrete, M., Saavedra-Molina, A., Campos-García, J., Valenzuela-Vázquez,U.,Amaya-Delgado,L.,&Arellano-Plaza,M.
   GenerationBioethanolProductionthroughaSimultaneousSaccharification-FermentationProcessUsingKluyveromycesMarxianusThermotolerant Yeast.Special Topics in Renewable Energy Systems(2018). doi:10.5772/intechopen.78052
- 45. MorakotKrajang;KwanruthaiMalairuang;JatupornSukna;KrongchanRattanapradit;SaethawatChamsart. Single-step ethanol production from raw cassava starch using a combination of raw starch hydrolysis and fermentation,scale-up from 5-L laboratory and 200-L pilot plant to 3000-Lindustrialfermenters.BiotechnologyforBiofuels(2021),doi:10.1186/s13068-021-01903-3.
- 46. Akihiko Kosugi; Akihiko Kondo; Mitsuyoshi Ueda; Yoshinori Murata; Pilanee Vaithanomsat; WaruneeThanapase;TakamitsuArai;YutakaMori.Productionofethanolfromcassavapulpviafermentation with a surface-engineered yeast strain displaying glucoamylase. 34(5), 1354–1358 (2009). doi:10.1016/j.renene.2008.09.002.
- 47. Papathoti, Narendra&Mendam, Kishore&Thepbandit, Wannaporn&Burgula, Niharika&Sangpueak, Rungthip & Saengchan, Chanon & Hoang, Nguyen & Keshav, Praveen & Le Thanh, Toan& Buensanteai, Natthiya. Bioethanol production from alkali-pretreated cassava stem waste viaconsolidatedbioprocessingbyethanoltolerantClostridiumthermocellumATCC31,924.BiomassConversionandBiorefinery (2022).10.1007/s13399-022-02868-5.
- 48. Wang,R.,Wang,D.,Gao,X.,Hong,J.Directfermentationofraw starch using aKluyveromyces marxianus strain that expresses glucoamylase and Alpha-amylase to produce ethanol.Biotechnol.Prog. 30(2), 338-347 (2014).