

Bioethanol (environment support fuel) production and optimization from pineapple peel and banana peel

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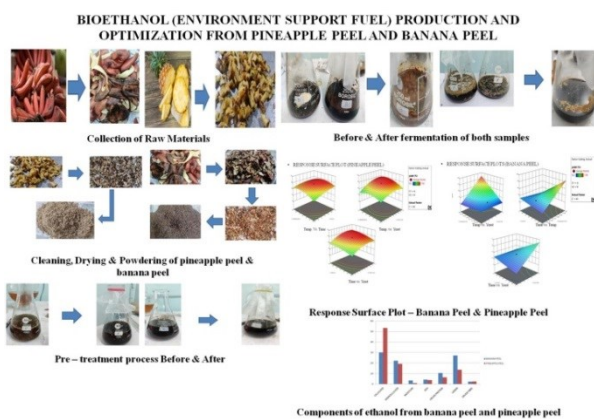
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Graphical abstract



Abstract

The goal of this investigation was to focus on the viability of synchronous aging of natural product waste with *Aspergillus Niger* TISTR 3063 co-societies producing ethanol. Additionally noted was how the aging temperature affected the yield of ethanol. Peels from pineapple and banana were used as substrates, and they were prepared by being slashed into little rectangular pieces. A 250 ml Erlenmeyer jar containing glucose was used to mature several bundles of natural product waste as a control. Analysis of the factors involved in the production of organic waste included sugar, pH, TS, vs, debris, moisture, COD, and TKN. In my review, bio ethanol was produced from banana strips and pineapple strips using pretreatment techniques, more specifically, soluble and acidic pretreatment. The process of aging interaction is then used to produce bioethanol. However, the production of bioethanol from food crops needs to be regulated to ensure food security and requires line-by-line analysis to move potential financial and environmental risks. Utilizing Plan Master Programming version 13, determine whether the nature of the bioethanol has changed after synthesis. Using plan master programming, it is possible to see how the item ethanol is being streamlined. Plot the chart for the item's actual and

predicted values, one component diagram, and 3D surface. The fuel qualities of the bioethanol that was given were thought to be comparable to those of diesel fuel, and they also complied with ASTM regulations.

Keywords: Pineapple peels, banana peels, *aspergillus niger* TISTR, simultaneous fermentation, ethanol, ethanol yield

1. Introduction

Ethanol has expected expanding significance as a biofuel as it very well may be delivered through maturation of different sorts of biomass. In Thailand, ethanol has been gradually eased in as an elective fuel by blending in with benzene to make a fuel known as gasohol. The Thailand Elective Energy Improvement Plan (2008-2022). Government policies and promotion measures for the "eco-ethanol" industry focus mainly at development of logistic systems to reduce cost of research, as well as identification of alternative sources of biomass as feedstock. Since use of squanders by means of aging offers a minimal expense elective for bio-ethanol creation, it is critical to evaluate the capability of elective wellsprings of biomass as substrate. Various investigations have bended on byproducts like vacant natural product packs from oil palm (Jung *et al.*, 2013).

Factors influencing ethanol yield in % incorporate pretreatment of the substrate; this is particularly significant for substrates containing high measures of cellulose. Corrosive or basic pretreatment utilizing either hydrochloric corrosive or watery alkali have been proposed, under various circumstances including steam cleansing, microwave and ultrasonic wave treatment (Garcia *et al.*, 2014). The utilization of enzymatic hydrolysis to speed up arrival of sugars has likewise been detailed. By the by, aging temperature is one of the most significant determinants of ethanol yield in %. Ideal maturation temperature has been broadly detailed in the scope of 30-38°C (Mishra and Ghosh, 2019). Moreover, the utilization of co-societies has too been concentrated form of further expanding ethanol yield in %. This gives an elective maturation way when biomass high in

lignocelluloses is utilized as feedstock (Wang *et al.*, 2019). For bio-squanders, co-societies of yeasts (*Saccharomyces cerevisiae* and *Pichia stipites*) accomplished ethanol focuses up to 45 g L⁻¹. As of not long ago, co-societies for concurrent saccharification and cofermentation utilized blends of microscopic organisms and yeast (Wang *et al.*, 2019), yeast and yeast (Ntaikou *et al.*, 2018) or growths and yeast (Izmirliloglu and Demirci, 2017) contingent upon substrate and aging circumstances. The utilization of co-societies not just expanded ethanol yield in %, be that as it may, can likewise diminish generally process costs.

Regardless of the various examination consideration on this subject, there stays extensive need to concentrate on the utility of different wellsprings of bio-squanders under different interaction conditions. The overhauling of squanders from organic product handlings a high-esteem substrate for bio-ethanol creation is specifically noteworthy in this respect, as a down to earth modern scale arrangement. Thusly, the target of this study was to concentrate on ethanol creation acquired from synchronous saccharification and cofermentation (SSCF) utilizing a co-culture of parasite and yeast. The impact of maturation temperature on ethanol creation was more over examined.

Pineapple juice extraction is finished on homegrown and modern level. The modern handling of natural product into juice and different items prompts the development of gigantic transient too as degradable waste (peels) to present grave dangerous effects if there should be an occurrence of ill-advised arranging off (Kanatt *et al.*, 2010). PPW is classifies as lignocelluloses squander materials, so it tends to be utilized to create bioethanol by weaken corrosive hydrolysis. This non eatable and biodegradable waste could supportenergy emergency by its bioconversion in ethanol and decreases the municipal squander (Khan *et al.*, 2015). A. Niger KL17, a yeast strain proficient in galactose use for ethanol creation was separated from soil tests and recognized through 18s RNA sequencing by Kim and partners (Azhar *et al.*, 2017). In the current study, BW was utilized as feedstock for bioethanol creation by. *Cerevisiae* KL17. The BW was saccharified utilizing acidic and enzymatic hydrolysis and the cycle was improved. The got sugars were matured into ethanol at first in shake flagon followed by increase in bioreactor in bunch and took care of group mode. Further to work on the interaction financial aspects, the buildups from ethanol aging and hydrolysis of BWwere pooled out and exposed to anaerobic assimilation (Promotion) to explore their Biochemical Methylation Potential (BMP). Afterward, the review was zeroed in on surveying the natural supportability through life cycle appraisal (LCA) and recognizing the effects of the different subprocesses and key ecological areas of interest that can direct future development and business (Murata *et al.*, 2015).

2. Materials and methods

2.1. Collection of banana peels and pineapple peels

Ripped banana peels and pineapple peels are collected from the market and fresh juice shops. After collecting the

peels, cut into small pieces by using knife and scissors. Then the peels are placed under the sunrays for drying from 5 to 6 days. After drying, with the help of mixer make them into powder form and mark as banana peel powder and pineapple peel powder (Figure 1).

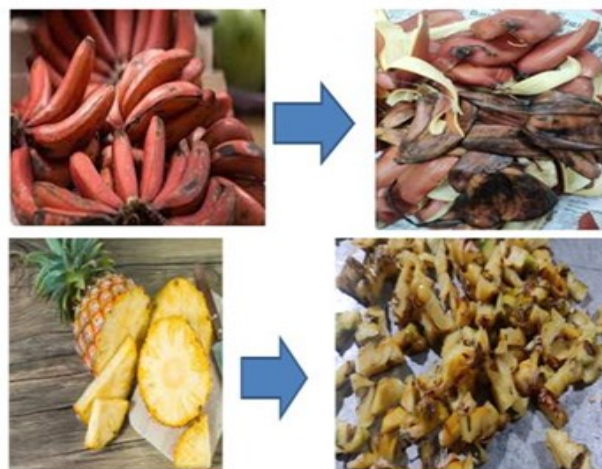


Figure 1. Collections of Raw Materials

2.2. Cleaning, drying and powdering

After collecting the pineapple peels and banana peel, clean them by using water latterly. After cleaning, cut the collected peels into smaller pieces, then the peels are dried in the sun and converted into powder by using mixer. The procedure is shown below (Figure 2).



Figure 2. Cleaning, Drying & Powdering of pineapple peel & banana peel

2.3. Chemical and physical treatment

15g of banana peel powder was added with 150 ml of distilled water and stir well for 2 minutes. After mixing 5g of potassium hydroxide is add into it and stir well up to 5 minutes.

15g of banana peel powder + 150ml of distilled water + 5g of KOH = pre-treatment solution

15g of pineapple peel powder was added with 150 ml of distilled water and stir well for 2 minutes. After mixing 5g

of potassium hydroxide is added into it and stirred well up to 5 minutes (Figure 3).

15g of pineapple peel powder + 150ml of distilled water + 5g of KOH = pre-treatment solution



Figure 3. Pre – treatment process Before & After

2.4. Solution for pretreatment

Pretreatment of lignocellulosic biomass preceding hydrolysis is an essential for bioethanol creation since it decides the yield of bioethanol that would be gotten after maturation. The point of pretreatment is to diminish the smallness, strength and glasslike nature of cellulose supporting hydrolyzing the lignocellulosic biomass to basic sugar units. The antacid pretreatment was done utilizing electrically warmed autoclave by the utilization of 10 % (wt/wt) KOH and alcohol to fiber proportion of 6:1. We cooked the fiber at 120°C for six hours before the release of strain into the environment. The pineapple and banana peels were washed with water and air-dried at 45 °C. We exposed the strips to water pretreatment as a pretreatment cycle in a shut autoclave. The pineapple and banana squander were cooked at 120 °C utilizing water to alcohol proportion of 1:10 for six hours. The tension was delivered into the environment and the pulped fiber was washed with water and air-dried at 45 °C. For corrosive pretreatment, 40 g of banana test was blended in with 200 mL of five percent H₂SO₄ and kept at 120 °C for six hours. The blend was sifted to isolate the strong buildups from the filtrate portion. The strong deposits were entirely washed with regular water to unbiased pH and dried at 45 °C. The production of bio ethanol was enhanced by alkaline pretreatment with NaOH under moderate working condition (Mirahmadi *et al.*, 2010). Same kind of alkaline pre-treatment was used for both samples with using KOH. After mixing of potassium hydroxide stir the solution up to 5 minutes, Heat the solution by using water tub and set the temperature of about 75°C for 2 hours and cooled it for 24 hrs.

2.5. Fermentation process

Before fermentation, filtered the pre-treatment sample to reduce the impurities present in the sample. With the help of hand kerchief filtered the sample 2 times. Add 15g of yeast Powder before fermentation process (Figure 4).

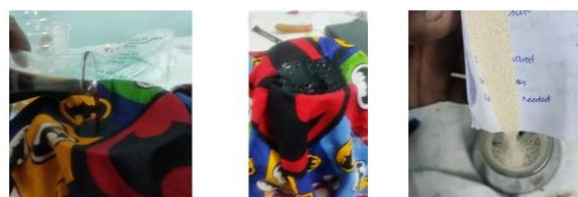


Figure 4. Filtration and yeast adding

Fermentation is the final stage in bioethanol production. We transformed the monosaccharides and some disaccharides produced during hydrolysis into ethanol using *S. cerevisiae* invertase and zymase enzymes (Figure 5).

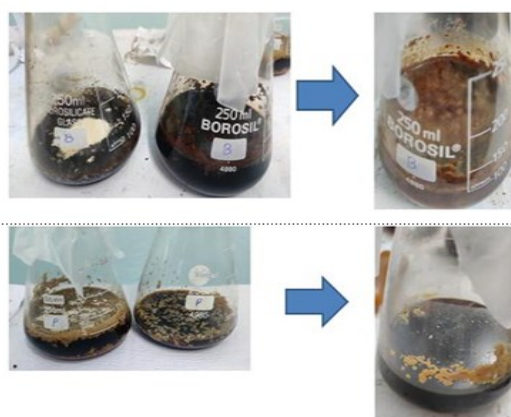


Figure 5. Before & After fermentation of both samples

The *S. cerevisiae* cells were suspended in deionized water, and the only carbon source for the yeast cells was processed pineapple and banana waste. Before and after fermentation using pineapple and banana peels, as well as yeast cells in deionized water. Fermentation is the process of sugars being broken down by enzymes of microorganisms in the absence of oxygen. For fermentation process, close the conical flask by using aluminum foil or plastic cover and keep it for 4 to 6 days for fermentation.

2.6. Statistical data analysis

The information acquired was dissected genuinely utilizing reaction surface technique. To fit the second-request mathematical model produced by the Plan Master programming form 10 (Detail Straight forwardness Inc., Minneapolis, USA), examination of fluctuation (ANOVA), a relapse investigation and the plotting of reaction surface were completed. From the numerical model developed, the trial information acquired from the laid out ideal condition were utilized as approving set, and these were contrasted and the anticipated qualities. The fitted second-request numerical model is depicted underneath.

2.7. Sugar concentration

Hydrolysis of pineapple and banana peels were completed at 2%, 4%, 6%, and 8% w/v substrate concentration (SC) using a commercial catalyst at a dosage of 10 U/g. DM for 72 hours. Hydrolysis often grew with increasing substrate

focus and increasing duration to an optimum and begins to reduce. The least amount of sugar was supplied at 2% substrate concentration after 12 hours (5.7 g/L and 4.5 g/L). Nonetheless, the concentration of sugars given at 8% SC was typically lower than that of 6% SC after 36 hours. From 36 to 72 hours, the 6% substrate concentration had the greatest enhanced sugar set free. Hydrolysis was comparable between 6% and 8% substrate focus after 24 hours ($p = 0.84$). It was also discovered that after 48 hours, there was no increase in sugars supplied across all substrate fixations. This is due to item constraint, when acids are given, lowering the pH of the medium and so reducing the compound's activity. Hydrolysis for pineapple peels and banana peels can thus be stopped after 48 hours to save money.

3. Result and discussion

$$Y = b_0 + \sum_{i=1}^k b_i X_i + \sum_{i=1}^k b_{ii} X_i^2 + \sum_{i < j}^k b_{ij} X_i X_j + e$$

ANNOVA Table 1. Banana peel ethanol

Source	Sum of Squares	df	Mean Square	F-value	p-value	
Model	11851.26	6	1975.21	3.7	0.0228	significant
A-yeast	3310.63	1	3310.63	6.21	0.0270	
B-temperature	1893.56	1	1893.56	3.55	0.0821	
C-time	394.33	1	394.33	0.7392	0.4055	
AB	2227.11	1	2227.11	4.17	0.0618	
AC	1949.38	1	1949.38	3.65	0.0782	
BC	2076.26	1	2076.26	3.89	0.0702	
Residual	6934.94	13	533.46			
Lack of Fit	6934.55	8	866.82	11160.76	< 0.0001	Significant
Pure Error	0.3883	5	0.0777			
Cor Total	18786.2	19				

These insights construed that the model created for the hydrolysis stage properly portrayed the genuine relationship among the picked factors. The p-upside of the model terms were critical at $p < 0.05$ (Table 4). Among the variables, the home time was not huge ($p > 0.05$). Furthermore, the noticed low p-worth of 0.0001 and the looking at high Fvalue of 143.24 in the sulphite fixation showed that the model obtained was huge. The difference between negative and positive tremendous impacts of each term in the model isn't shown by both the F-worth and p-esteem as detailed by Betiku and Taiwo (2015). The information in regard to coded factors relating the glucose yield (kgm-3) Y with free factors were fitted utilizing the next second-request polynomial condition.

The model terms (A, B, Stomach muscle, B2, A2, C,2 AC, BC) with positive coefficients show a decent effect on wipe gourd hydrolysis while the model terms (C) with negative coefficient illustrate negative fit on wipe gourd hydrolysis. Yield \times Temperature (cross result of the straight term) was the best critical model term on wipe gourd hydrolysis followed by sulphite focus, Temperature (quadratic term), and Temperature (straight term). This perception is confirmed by the p-values (Table 4) of these model terms ($p > 0.05$). The result

The trial results got above was inputted into the exploratory plan produced by RSM to acquire the investigation of difference (ANOVA) of the relapse condition model showed. A quadratic model was proposed as showed upon Table 1. F-worth of 214.49 with a low likelihood worth of less than 0.0001 infers that the model is critical. Blend of different coefficients of connection (R) and coefficient of assurance (R^2) was used to study the integrity of attack of the relapse condition. R of 0.9987 of the models showed decent relationship between's the noticed and anticipated values. While R^2 of the model was 0.9974, which illustrated that 99.74% example variety for glucose delivered is inferable from the free factors and only 0.26% of the aggregate varieties are not portrayed by the model. The changed R^2 of 0.9928 exhibited that the model was huge. It has been recommended that R^2 should be between 80-100 percent for the solid match of a model.

showed that the cross result of straight term of Temperature and Response time contributed the least to the hydrolysis of Wipe gourd. The low assessment of the standard blunder of the capture and each model term uncovered that the relapse model fits the information well and the expectation was perfect. The difference expansion factor (VIF) acquired in this work uncovered that the focus focuses are symmetrical to each and every model. This is as per the review that Betiku and Taiwo (2015) led on yam. Indeed, the elevated degrees of the variables are coded as +1 and low levels of the elements are coded -1.

The coded condition is significant for perceiving the general impact of the elements by checking out at the coefficients of the variables. Notwithstanding, as far as real factors, the levels should be determined in the first units for every one of the elements. The condition as far as genuine elements can't be used to choose the overall commitment of each component in light of the fact that the coefficients are scaled to suits the units of each element and capture isn't at the focal point of the plan space Equation (Tables 2 and 3).

ANNOVA Table 2. For pineapple peel ethanol

Source	Sum of Squares	df	Mean Square	F-value	p-value	
Model	1164.04	9	129.34	12.83	0.0002	Significant
A-YEAST	372.81	1	372.81	36.99	0.0001	
B-TIME	124.78	1	124.78	12.38	0.0056	
C-TEMPERATURE	46.55	1	46.55	4.62	0.0572	
AB	2.42	1	2.42	0.2401	0.6347	
AC	36.98	1	36.98	3.67	0.0844	
BC	16.82	1	16.82	1.67	0.2255	
A ²	308.54	1	308.54	30.61	0.0003	
B ²	52.28	1	52.28	5.19	0.046	
C ²	292.26	1	292.26	29	0.0003	
Residual	100.79	10	10.08			
Lack of Fit	100.68	5	20.14	888.32	< 0.0001	Significant
Pure Error	0.1133	5	0.0227			
Cor Total	1264.83	19				

Michaelis-Menten motor model for enzymatic hydrolysis and maturation was exposed to mistake investigation, utilizing three measurable apparatuses, and mean square mistake (RMSE), change and outright normal rate deviation (AAD), to assess the proficiency of the model with the exploratory information. Measurable investigation utilizing RMSE, difference and AAD was researched. Typically, the higher the worth of the R2 and bring down the upsides of the RMSE, the better would be the integrity of the model to fit the trial information.

• RESPONSE SURFACE PLOT (PINEAPPLE PEEL)

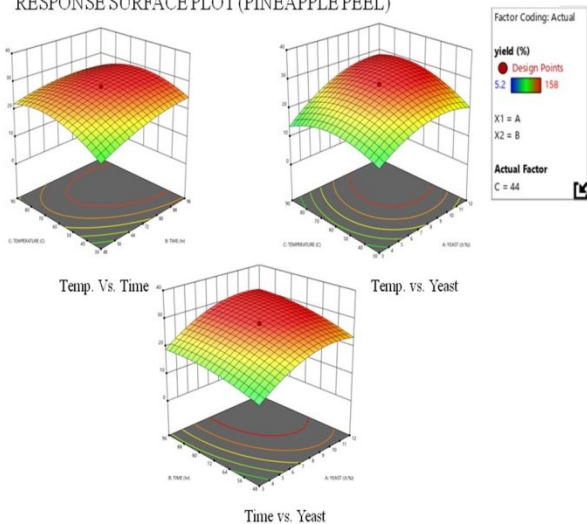


Figure 6. Response Surface Plot – Pineapple Peel

The RMSE is a habitually involved measurement in looking to information, anticipated and noticed values by a model. The RMSE could be applied to confirm exploratory outcomes, got in the investigation of enzymatic hydrolysis and aging active models. This can be accomplished by showing the distinctions between the qualities anticipated by a model and the qualities noticed. Thus, the RMSE stays a commendable proportion of exactness. It is likewise the square foundation of fluctuation (σ^2). Nonetheless, lesser upsides of σ^2 , RMSE and AAD are by and large, better than higher qualities. From Figures 6

and 7, a quantitative proportion of execution was assessed.

• RESPONSE SURFACE PLOTS (BANANA PEEL)

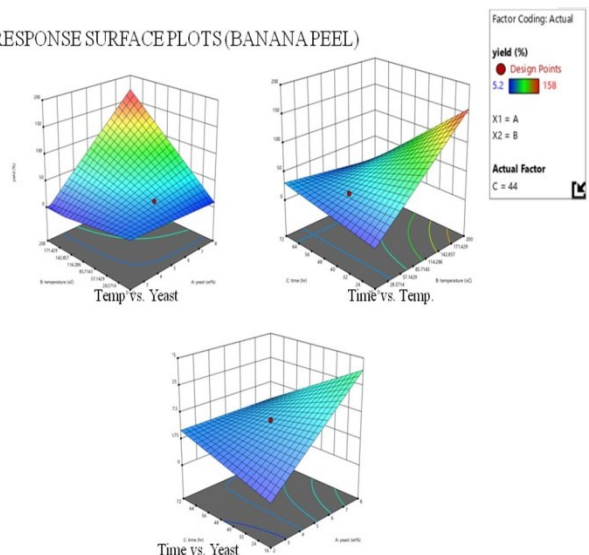


Figure 7. Response Surface Plot – Pineapple Peel

A quadratic model was chosen, and the model was critical. The model Fisher F-trial of 15.25 with low likelihood esteem [(model > F) =0.0002] exhibit a high importance for the relapse model (Ademakinwa et al., 2019). There was just a 0.02 % chance that an F-esteem this enormous was not critical, which could happen because of commotion. Sufficient accuracy estimates the sign to clamor proportion.

A proportion more prominent than 4 is attractive, the sufficient accuracy is 13.395 which shows a satisfactory sign. This model can be utilized to explore the plan space. The outcomes showed that the p-up sides of the model terms were huge, i.e., $p < 0.05$. The more modest the extent of P-esteems, the more critical is the comparing coefficient (Joglekar and May, 1987). For this situation, the two direct terms (B, C), the quadratic term (A2, B2, AC) are huge model terms at 95% certainty level.

The integrity of fit was actually taken a look at by the coefficient of assurance (R2). For this stage, the R2 was at

93.13 % and the changed R2 was viewed as 87.02 %, the sensible arrangement between the R2 and changed R2 suggested that in the model examined, that they chose factors are appropriate for the model (Ross, n.d.). The worth of the R2 acquired from this work showed a high consistency between the noticed qualities and the anticipated qualities. These qualities showed that the relapse is genuinely huge; just 0.02 % of the complete variety was not made sense of by this relapse model. The low upsides of standard mistake saw in the catch and every one of the model terms showed that relapse model fits the information appropriately and that the forecast was great. The change expansion factor (VIF) shows that the middle focuses are orthogonal to all other factors. The data were fitted using the following second-order polynomial equation.

$$Y = 5.89 + 0.051A - 0.22B + 0.43C - 0.030AB + 0.25AC + 0.017BC - 0.91A^2 + 0.31B^2 \quad (1)$$

Where Y is the bioethanol delivered in kgm^{-3} and is inoculums size in v/v, B is maturation time, C is nitrogen source. This condition can be utilized to make expectations of ethanol yield in % under shifting states of the specified variables of aging time, inoculums size and nitrogen source. Naturally, the elevated degrees of the variables are coded as +1 and Low levels of the variables are coded -1. The coded condition is helpful for distinguishing the general effect of the variables by looking at the coefficients of the elements. RSM has demonstrated to be a worthwhile instrument for improving society conditions and Culture media creation (Betiku and Taiwo, 2019; Bezerra *et al.*, 2008).

4. Process optimization of fermentation stage

The ideal upsides of the free factors chose for the maturation step were additionally acquired by tackling and utilizing the Plan - master programming. The ideal condition was researched to be A= 7.500, B=24.000 and C=inorganic. The ethanol fixation anticipated under the

above set of condition was 6.840 kgm^{-3} . To confirm the forecast of the model, the ideal condition was run on two free reproduces and the typical ethanol concentration was 6.632 kgm^{-3} , which is well inside the anticipated worth of the model condition. The consequences of this work show that the reaction surface technique could be utilized to advance ethanol creation yield from wipe gourd. The outcomes gotten in this exploration for bioethanol creation from wipe gourd are along these lines promising (Rorke and Gueguim Kana, 2017; Ballesteros *et al.*, 2004, Fan *et al.*, 1987). The ethanol yield in % decreased up to the mid-range concentration, followed by a further increase in the yield. Also, from the plot bioethanol yield for the inorganic nitrogen source is higher than when the nitrogen source is organic for all fermentation time. After producing ethanol from banana peel and pineapple peel, test both samples in the PRECISE ANALYTICS lab in Arumbakkam, Chennai (Figure 8).

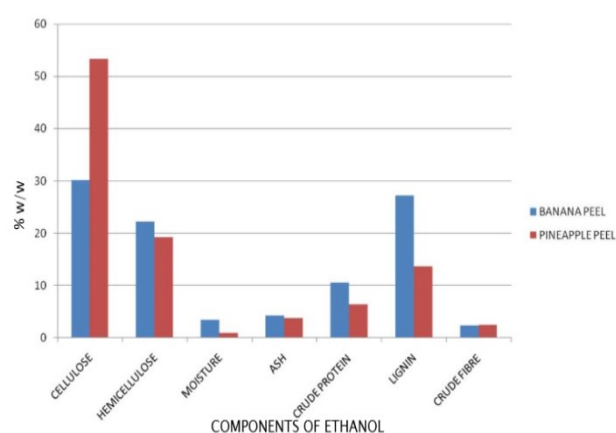


Figure 8. Comparison chart between the components of ethanol from banana peel and pineapple peel

The test report are given below.

Table 3. Comparison results for Ethanol production from Banana Peel and Pineapple Peel

Components	Ethanol from Banana Peel	Ethanol from Pineapple Peel
	% w/w	% w/w
Cellulose	30.25	53.36
Hemicellulose	22.24	19.34
Moisture	3.6	1.0
Ash	4.34	3.76
Crude Protein	10.51	6.54
Lignin	27.21	13.62
Crude Fibre	2.4	2.52

It has been demonstrated that simultaneous saccharification and fermentation may efficiently remove glucose, a cellulase inhibitor, increasing the yield and rate of cellulose breakdown. Despite the fact that ethanol is created from renewable resources, economic considerations such as land availability, labor, taxation, utilities, agricultural processing charges, and transportation must be taken into account; otherwise, there will be no profit from its production.

The cost of raw materials has a considerable impact on the cost of bioethanol production, accounting for 40-75% of total expenses depending on the kind of feedstock. Bioethanol manufactured from pineapple peel and banana peel offers minimal manufacturing costs in India. The cost of manufacturing bioethanol can be significantly compensated by lowering greenhouse gas emissions, assuring energy supply security, and increasing rural agricultural activities. The most challenging challenge is

decreasing the cost of producing bioethanol. As a result, the biorefinery concept is required to make greater use of renewable feedstocks (Pineapple Peel and Banana Peel) and to generate additional value-added coproducts (e.g. bio-based materials from lignin) that would reduce the cost of bioethanol production.

5. Conclusion

Reaction surface technique was effectively used to upgrade the enzymatic hydrolysis and maturation stages. The mixes that gave the ideal glucose yield during the hydrolysis are yeast, time & Temperature (100 °C) and Response Time (60 mins) with a glucose concentration of 6.673 kgm⁻³. For the aging step, the ideal condition was explored as 7.5 % v/v inoculum size; maturation season of 24 h; and inorganic Nitrogen source using A. Niger with an ethanol fixation of 6.84 kgm⁻³. It very well may be closed from this study that waste gourd (a lignocelluloses material) can be a decent feedstock for bioethanol creation and could forestall wastage of underutilized agro material. The purpose of the present work is to investigate the possibility using banana & pineapple peels are to be more cost effective. Therefore, this report emphasizes that enzymatic hydrolysis of the peels are easily fermented by yeast. In conclusion, the data obtained for fermentation in lignocelluloses residues revealed that enzymatic hydrolysis indicates that the peels are stands as an alternative feed stock and is economically favorable for bioethanol production.

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