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Valorization of Unripe Plantain (*Musa paradisiaca*) Peel for a Renewable Bioethanol Production

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Abstract: The increasing demand for sustainable alternatives to fossil fuels has stimulated interest in lignocellulosic biomass as a renewable feedstock for bioethanol production. This study assessed the potential of unripe plantain (*Musa paradisiaca*) peels as a lignocellulosic substrate for bioethanol production, with emphasis on physicochemical composition, reducing sugar recovery, inhibitor reduction and ethanol yield. Fresh unripe plantain peels were washed, sundried, milled and subjected to combined pretreatment using a combination of 10% formic acid and 5% tetraoxosulphate vi acid at 121ºC for 30min, followed by acid hydrolysis. The physicochemical composition of the raw and pretreated biomass were determine using standard methods. The raw unripe plantain peel contained 54.34% carbohydrate, 28.60 cellulose, 17.40% hemicelluloses and 13.80% lignin. Following acid pretreatment, cellulose increased to 48.63.%, while hemicelluloses and lignin decreased to 8.63% and 7.46% respectively, indicating effective disruption of the biomass structure. The acid hydrolysis of the pretreated biomass produced a hydrolysate with 21.59g/l reducing sugars, corresponding to 51.80% sugar recovery. Detoxification of the hydrolysate using overliming and activated at 1:3 and 3:1 ratios reduces the inhibitor index by 40,98% and 54.76% respectively, with 3:1 ratio showing greater detoxification efficiency. Fermentation of the hydrolysate using yeast produced an ethanol concentration of 3.82g and 14.86% yield for undetoxified sample, 6.43g and 38.91% yield at 1:3 ratio, and 8.42g and 50.18% at 3:1 detoxification ratio. The findings demonstrate that unripe plantain peels possesses considerable potential as a renewable lignocellulosic feedstock for bioethanol production. Valorization of unripe plantain peel waste contributes to waste management, renewable energy generation, and sustainable biofuel development.

Key words: Lignocellulosic biomass, Renewable energy, unripe plantain peels, Fossil fuel, Bioethanol

1. INTRODUCTION

The increasing demand for energy and the environmental consequences associated with fossil fuel consumption have intensified the search for sustainable and renewable energy alternatives. Bioethanol is considered a promising renewable biofuel because it can be produced from biomass and used as a substitutes for or blended with gasoline. However the use of edible crops such as maize, sugarcane, and cassava for ethanol production has raised concerns regarding competition between the production and food supply. Consequently, increasing attention is being directed towards agriculture residues and other non-food biomass are inexpensive and sustainable feedstocks for second generation bioethanol (Kazmi et al., 2024; Niu et al., 2024).

Agricultural residues are particularly attractive for bioethanol production because they contain structural carbohydrates, mainly cellulose, hemicelluloses, which can be converted into fermentable sugars and subsequently fermented to ethanol. Their utilization also provides an effective strategy for reducing agricultural waste while generating value – added products. The conversion of lignocellulosic residues into biofuels is therefore consistent with circular-economy principles, in which waste materials are transformed into useful resources rather than being discarded (Mujtaba et al., 2023; Shahzad et al., 2024). Nevertheless, the complex association between cellulose, hemicelluloses and lignin makes lignocellulosic biomass resistant to direct hydrolysis, necessary suitable processing to improve carbohydrate accessibility and sugar recovery (Wang et al., 2024). Recent research indicates that appropriate pretreatment can improve sugar recovery and bioethanol yield, whereas sever treatment conditions may cause sugar degradation and generate fermentation inhibitors such as furfural, hydroxylmethylfurfural (HMF), and phenolic compounds (Charaborty et al., 2024; Wang et al., 2024).

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Plantain (*Musa spp*) is widely cultivated and consumed in tropical regions, including Nigeria, generating substantial quantities of peel as an agricultural and household residue. Plantain peel is often discarded to decompose despite containing appreciable amount of carbohydrate and lignocellulosic components. Its conversion into bioethanol could therefore provide a sustainable approach for managing plantain waste while producing renewable energy. Recent studies have demonstrated the feasibility of converting plantain peels into bioethanol. Ajiboye et al., (2025) investigated bioethanol production from unripe plantain peels through fermentation using saccharomyces cerevisiae and Aspergillus niger. Similarly, Jumare et al., (2022) investigated bioethanol production from unripe plantain peel using S. cerevisiae and demonstrated its potential as a substrate for ethanol generation.

Despite this potential, efficient conversion of unripe plantain peel to ethanol depends on effective disruption of its lignocellulosic structure, release of fermentable sugars and subsequent microbial fermentation. Processing condition must also be carefully controlled because excessive treatment may result in sugar degradation and formation of compounds that inhibit fermentation (Niu et al., 2024; Wang et al., 2024). Valorizing this readily available agricultural residue could simultaneously contribute to waste reduction, renewable-energy generation and the development of non-food based biofuel resources.

II. MATERIALS AND METHODOLOGY

a. Sample Collection

Fresh mature unripe plantain peels were collected from local markets and household processing point in Dakingari, Kebbi State. The samples were placed in clean polyethylene bags, and immediately processed to minimized deterioration.

b. Sample Treatment

The collected peel were thoroughly washed with water to remove dirt and impurities that could interfere with chemical composition and subsequently rinsed with distilled water. The cleaned samples were sun-dried to constant weight. Then the dried peel was grounded with pestle and mortal to obtain a fine powder to enhance surface area and improve further processing. The powdered samples were sieved to obtained a fine particles. This was stored in a clean, airtight container at room temperature prior to physicochemical analysis (Fan *et al*., 2025).

c. Physico-Chemical Composition of Unripe Plantain peels

The physicochemical composition of the unripe plantain was determined using standard analytical methods. Moisture content was determined by oven-drying a known weight of the powdered sample to constant weight, while the ash content was determined by incineration in a muffle furnace. Crude fibre was determined using the standard fibre-analysis procedure, carbohydrate content was calculated by difference using the equation: Carbohydrate (%) = 100-(moisture + ash + protein + fat + fibre). All analysis were performed in triplicate and expressed as mean ± standard deviation. The analytical procedures were based on standard methods reported for unripe plantain peel characterization (Umoh, 2024).

The determination of these components is important for evaluating the suitability of unripe plantain peel as a bioethanol feedstock. In particular, its carbohydrate and fibre fractions provide an indication of the availability of structural and fermentable carbohydrates for subsequent conversion.

d. Determination of Lignocellulosic Composition of Unripe Plantain peel

The cellulose, hemicelluloses and lignin contents of the unripe plantain peel were determined using a standard gravimetric method. The dried sample was subjected to sequential chemical treatment, to remove soluble extractives and isolate the structural fibre fractions. The resulting residues were filtered, washed, dried to constant weight and weighed. The cellulose, hemicelluloses, and lignin fractions were determined from the differences in the weights of the residues obtained after the respective treatment. The contents of the individual lignocellulosic components of the individual lignocellulosic components were calculated as percentages of the initial dry sample weight.

e. Acid Pretreatment of unripe plantain peel

The dried unripe plantain peel was pretreated using combination of 0.8% formic acid and sulphuric acid. The mixture was prepared in the ratio 10%/5%. Approximately 6.0g of the oven-dried mango peels was weighed into three different conical flask and 60ml each of the ratio was added in ratio 1:10 (w/v). The mixture was thoroughly mixed and

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subjected to pretreatment in an autoclave at 121ºC for 30mins. After pretreatment, the reactor was allowed to cool to room temperature. The slurry was filtered to separate the liquid hydrolysates and the solid residue. The solid fraction was washed repeatedly with distilled water until a neutral pH was attained (Li *et al.*, 2024; Chen *et al.*, 2025). The solid residue was oven-dried and the chemical composition of the pretreated unripe plantain peels was determined. The recovered solid residue was washed several times with distill water until approximately neutral pH was obtained and was subsequently used for hydrolysis. The liquid fraction was collected for determination of reducing sugars and fermentation inhibitors.

f. Determination of Reducing Sugar in the hydrolyzates

The reducing sugar content of the hydrolysate was determined using the 3,5-dinitrosalicylic acid (DNS) colorimetric method, following the procedure reported by Khaire et al., (2025). The method is based on the reduction of DNS by reducing sugars under alkali heating conditions, producing a coloured product measurable at 540nm.

The DNS reagent was prepared by dissolving 1.0g of 3,5-dinitrosalicylic acid in distilled water, followed by the addition of sodium hydroxide and sodium potassium tartrate. The solution was thoroughly mixed until complete dissolution and made up to the required volume with distilled water

Analytical Procedure

The sugar hydrolyzates 1ml (50µ) was transferred into cleaned dried test tube and1ml (50µ) of DNS solution was added and mixed thoroughly. These were heated in a water bath at 100 ºC for 10min to allow complete colour development. The text tubes were allowed to cooled to room temperature, after which 5ml of distilled water was added to stabilized the colour. The concentration of the reducing sugars was determined by measuring the absorbance at 540nm using UV-Visible Spectrophotometer (Khaire et al., 2025).

g. Detoxification of the Fermentation Inhibitors in the Sugar Hydrolyzates

The acid hydrolysis was detoxified using combined overliming and activated charcoal in the ratio 1g/3g and 3g/1g (w/w) . The detoxification of the acid hydrolyzates employed the method of (Adewuyi *et al*., 2024; Liu *et al.*, 2025).

An approximately 5ml of the hydrolyzates of 0.8% (10%/5% v/v) was transferred into a clean 100ml beakers. To the hydrolyzates, 3g of the overliming was added to the hydrolyzates under a continous stirring until a pH of 10 is achieved. The mixture was heated in a water bath at a temperature of 60 ºC for 45min to facilitate chemical transformation and precipitation of inhibitory compounds.

Without removing the precipitates formed during overliming, 1g of the activated charcoal was directly added to the reaction mixture. The suspension was heated in a water bath and continuously stirred at the same temperature for additional 30min to allow simultaneous adsorption of residual inhibitors.

Following the combined treatment, the mixture was allowed to cooled to room temperature and subsequently centrifuge at 5000rpm for 15min to remove the calcium salts, degraded inhibitor complexes, and spent activated charcoal. The supernant was filtered and the filtrate was adjusted to pH of 5 using dilute sulphuric acid to create a favourable conditions for yeast fermentation. The detoxified hydrolyzates was stored prior to further analysis and fermentation processes. This was repeated for the other combination ratio of overliming/activated charcoal on the hydrolyzates. The percentage of inhibitors reduction can be calculated as

IR % = 1 - IAT/IBT X 100.

Where IR = percentage of inhibitors reduction , IAT = Inhibitors after treatment, IBT = Inhibitors before treatment.

h. Quantification of Degradation Inhibitors Using UV-Visible Spectrophotometry

Approximately 10ml of the detoxified hydrolyzates were transferred into a tube and centrifuge at 8000 rpm for 10min to remove suspended solids followed by filtration to eliminate fine particles and insoluble residues that could interfere with absorbance measurement. The pH of the hydrolyzates was adjusted to 5 using dilute hydrochloric acid to ensure spectra stability of furans and phenolic compound (Li *et al.*, 2023). The hydrolyzates was treated with activated charcoal toabsorb high-molecular weight lignin fragments, centrifuge, and then decant to get clear supernantant for analysis. The UV-Visible analysis was conducted using a spectrophotometer with distlled was the blank. Absorbance was recorded at 277nm of the estimation of furfural, 284nm for the HMF, and 280nm for phenolic compound.

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i. Fermentation of the unripe plantain peel Hydrolyzates Using Yeast

Fermentation is a biochemical conversion wherein fermentable sugars present in lignocellulosic hydrolyzates are transformed into ethanol and carbondioxide through the metabolic activity of yeast, Saccharomyces cerevisiae is the most commonly employed microorganism due to its high tolerance to ethanol, rapid glucose metabolism, and well-characterized fermentation pathways.

The hydrolyzates 20ml was transferred into 100ml conical flask. The pH of the hydrolyzates was adjusted to 5.5. Then the flask was inoculated with 2% (w/v) of the yeast. The flask was then incubated anaerobically at 30^0C to 35^0C for120hrs. Upon completion of the fermentation, the yeast cells was separated from the fermented hydrolyzates. Theses were subjected to 2% (w/w) of calcium chloride, agitate continuously for 10min to remove the water present in the fermented broth. The supernant was collected for quantification of ethanol and further analysis.

III RESULTS AND DISCUSSION

Table 1 shows that the raw unripe plantain peel contained appreciable carbohydrate (54.53%) and crude fibre (13.46%), indicating its suitability as a lignocellulosic feedstock for bioethanol production. After acid pretreatment, carbohydrate increased to 67.82%, while crude fibre, protein, ash, and fat decreased. The increase in carbohydrate may be associated with the preferential solubilisation of non-carbohydrate components during pretreatment. Such compositional changes are consistent with recent studies reporting that chemical pretreatment disrupts lignin-hemicellulose associations and improves accessibility of structural carbohydrates for subsequent hydrolysis (Chakraborty et al., 2024); Abolore et al., 2024). Similarly, recent reports also revealed that plantain peel contains substantial carbohydrate and structural fibre, supporting its valorization as a renewable biofuel feedstock (Akinola et al., 2025).

Table 1 : Physico-Chemical Composition unripe plantain (*Musa paradisiaca*) Peels

Parameters	Moisture	Ash Content	Crude fibre	Crude protein	Crude fat	Carbohydrate
Raw(%)	8.42 ±0.18	11.86 ±0.24	13.46 ±0.42	6.72 ±0.31	4.94 ± 0.31	54.53 ±0.76
10%FA/ 5%H_2SO_4 (%)	10.22 ±0.16	8.74 ±0.21	7.35 ±0.15	5.65 ±0.27	3.92 ±0.14	67.82 ±0.17

Table 2 indicates that acid pretreatment substantially alter the lignocellulosic composition of unripe plantain peel. Cellulose increased from 28.60% in the raw biomass to 48.72% after pretreatment, representing enrichment of the cellulose fraction. In contrast, hemicelluloses decreased from 17.40% to 8.63%, while lignin decline from 13.80% to 7.46%. These changes suggest that the combined acid treatment effectively solubilized hemicelluloses and lignin fractions and increased cellulose accessibility for hydrolysis. Similar changes were reported where removal of non-cellulosic components enhances carbohydrate accessibility and fermentable sugar release (Li et al., 2024; Chen et al., 2025).

Table 2 : Lignocellulosic Composition unripe plantain (*Musa paradisiaca*) Peels

Parameters	Cellulose	Hemicellulose	Lignin
Raw(%)	28.60 ±0.65	17.40 ±0.43	13.80 ±0.37
10%FA/ 5%H_2SO_4 (%)	48.72 ±0.91	8.63 ±0.37	7.46 ±0.29

Table 3 : Determination of Reducing Sugars of unripe plantain (*Musa spp*) Peels hydrolysates

Absorbance of Standard = 0.872nm

Absorbance of Blank =0.071nm

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Parameters	Carbohydrate Content	Vol. of hydrolysate (cm^3)	Abs. of spl (nm)	Reducing Sugar (g/l)	Red.S. Recovery (%)
10%FA/H_2SO_4 (%)	67.82	27.69	0.82	21.59	51.80

Table 3 presents the reducing sugar content obtained after acid hydrolysis of pretreated unripe plantain peel biomass, produced 21.65g/l reducing sugars, indicating effective conversion of structural carbohydrates into fermentable sugars. The value is comparable with the recent reports of approximately 19.35-26.42g/l reducing sugars from pretreated unripe plantain peel. The results confirms suitability of acid pretreatment for enhancing sugar release from unripe plantain biomass (Jayamma et al., 2025). The 51.80% recovery observed in this study indicates that the combined acid pretreatment effectively disrupt the lignocellulosic matrix and made carbohydrate fraction accessible for hydrolysis. Ajiboye et al., (2025)obtained higher sugar concentrations from unripe plantain peels using enzymatic hydrolysis.

Table 4 : Quantification of inhibitors in unripe plantain (*Musa spp*) Peels hydrolysate

Detoxification Ratio	A_{277}	A_{280}	A_{284}	A_{320}	$A_{(\text{index } 280\text{-}320)}$	% Reduction
Before	0.743	0.796	0.772	0.450	0.346	-
1:3	0.520	0.584	0.470	0.371	0.213	40.96
3:1	0.365	0.382	0.224	0.196	0.186	54.76

Acid hydrolysis of lignocellulosic biomass generates fermentation inhibitors such as furfural, HMF, and phenolic compounds that absorb at 277nm, 284nm, and 280nm respectively. These compounds are toxic to yeast and can significantly reduce ethanol yield.

The results in Table 4 shows that detoxification subsequently reduced the inhibitor index of the unripe plantain peel hydrolysate. The 1:3 overliming/activated charcoal ratio reduced inhibitors by 40.96%, whereas the 3:1 ratio achieved 54.76% reduction, indicating greater detoxification efficiency. The superior performance of the 3:1 treatment may reflect complementary chemical transformation and adsorption of inhibitory compounds. Similar studies reports effective removal of furfural, HMF, and phenolic compounds using combined overliming and activated charcoal treatment, thereby improving hydrolysate fermentability (Zhang et al., 2024) and Liu et al., (2025) demonstrated that overliming at pH10 followed by activated charcoal treatment improves hydrolysate fermentability by detoxifying inhibitors without major sugar loss. The higher percentage reduction at 3:1 ratio also correlates with the ethanol yield in Table 5, where the 3:1 detoxified sample gave the highest ethanol concentrations of 8.42g and yield of 50.18%. This confirms that effective detoxification directly improves fermentation performance.

Table 5: Determination of bioethanol from the unripe plantain (*Musa spp*) peel hydrolysate

S/N	Pretreatment (%)	Detoxification Ratio	Ethanol concentration (g)	Ethanol yield (%)
1	10%FA/5%H_2SO_4	Undetoxified	3.82	14.86
2	10%FA/5%H_2SO_4	1:3	6.43	38.91
3	10%FA/5%H_2SO_4	3:1	8.42	50.18

From the results obtained during fermentation as shown in table 5, demonstrate that detoxification enhanced bioethanol production from unripe plaintain peel hydrolysate. Ethanol yield increased from 14.86% in undetoxified hydrolysate to 38.91% and 50.18% following 1:3 and 3:1 detoxification, respectively. The higher yield at 3:1 corresponds with improved yeast fermentation. Similar studies confirm the potential of unripe plantain peel as a substrate for ethanol production.

This trend directly correlates with the inhibitor reduction data in Table 4. The 3:1 ratio achieved 64.76% inhibitor reduction compared to 45.20% for the 1:3 ratio. Lower inhibitor levels reduces stress on yeast during fermentation, allowing more efficient conversion of sugars to ethanol. This supports findings by Zhang et al., (2024) and Adewuyi et al., (2024) that combined overliming and activated charcoal detoxification enhances fermentability of lignocellulosic hydrolysates.

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IV CONCLUSION AND RECOMMENDATIONS

The study assessed the potential of unripe plantain (*Musa paradisiaca*) as a lignocellulosic feedstock for renewable bioethanol production. Acid pretreatment improved the relative cellulose content while reducing hemicelluloses and lignin. Hydrolysis generated appreciable reducing sugars, while combined overliming and activated charcoal effectively reduced fermentation inhibitors. The 3;1 detoxification ratio produced the highest inhibitor reduction and ethanol yield, confirming that effective detoxification enhances fermentability and bioethanol production from unripe plantain peel hydrolysate. The findings confirm that unripe plantain peel, an abundant agricultural waste in Nigeria, has considerable potential for bioethanol production. Effective pretreatment and detoxification, are essential for maximizing sugar release and fermentation efficiency.

It is recommended that further studies should optimize the pretreatment and hydrolysis conditions of unripe plantain peel to maximize fermentable sugar recovery while minimizing sugar degradation and inhibitor formation. Alternative pretreatment methods, including Alkaline, enzymatic and combined approaches, should also be evaluated to improve delignification and hydrolysis efficiency. The use of efficient and inhibitor-tolerant microorganisms should be investigated to enhanced ethanol fermentation. Furthermore, integrated biorefinery approaches should also be explored to recover ethanol and other valuable products, thereby improving wastes utilization and supporting sustainable bioenergy production.

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