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Biological Valorization of Banana Pseudostem through Biological Conversion: HPLC, GC and FTIR Characterization

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ABSTRACT

Banana pseudostem is an abundant agricultural residue containing substantial lignocellulosic carbohydrate and represents a promising renewable biomass resource for value-added bioproduct development. The present study investigated the biological conversion of banana pseudostem (BPS) followed by fermentation and characterization of the resulting product using complementary analytical techniques. The carbohydrate-containing fraction obtained after biological treatment was analyzed by high-performance liquid chromatography (HPLC), which showed a prominent peak corresponding to glucose. The glucose-containing fraction was subsequently subjected to yeast fermentation, and the resulting product was characterized by gas chromatography (GC) using an ethanol reference standard and by Fourier-transform infrared spectroscopy (FTIR). Comparison of the GC chromatograms showed a prominent sample peak at a chromatographic position corresponding closely to the major peak of the ethanol standard, providing evidence consistent with the presence of ethanol in the fermented product. The FTIR spectrum exhibited a broad O–H stretching absorption together with characteristic aliphatic C–H and C–O absorption features consistent with an alcohol-containing compound. The agreement among the HPLC, GC and FTIR observations supports the proposed conversion of banana pseudostem-derived carbohydrate into an ethanol-containing fermentation product. However, the present qualitative analytical data do not establish ethanol concentration, fermentation efficiency, yield, or absolute product purity. The study demonstrates the analytical feasibility of investigating banana pseudostem as a lignocellulosic agricultural residue and contributes to the broader development of biomass-valorization strategies.

Keywords: banana pseudostem; lignocellulosic biomass; biological conversion; glucose; fermentation; ethanol-containing product; HPLC; gas chromatography; FTIR; biomass valorization.

1. INTRODUCTION

Lignocellulosic agricultural residues are increasingly being investigated as renewable feedstocks for the production of value-added chemicals and biofuels. Such biomass mainly consists of cellulose, hemicellulose and lignin, and the close association among these components makes the carbohydrate fraction relatively resistant to enzymatic hydrolysis. Appropriate pretreatment or biological treatment is therefore generally required to improve accessibility of cellulose and hemicellulose to hydrolytic enzymes [1,2].

Lignocellulosic biomass represents an attractive alternative to food-based feedstocks for second-generation bioethanol production because agricultural residues can serve as carbohydrate sources without directly competing with food resources [3,4]. Banana pseudostem is particularly interesting because it is generated in large quantities as a residue of banana cultivation and contains appreciable amounts of structural carbohydrates. A recent review of banana wastes identified banana pseudostem as one of the particularly promising banana residues for bioethanol production [5].

Several studies have demonstrated the feasibility of utilizing banana pseudostem for ethanol production. Ingale et al. reported the production of bioethanol from banana pseudostem following pretreatment and microbial treatment, demonstrating the potential of this agricultural residue as a substrate for ethanol production [6]. Similarly, Legodi et al. investigated enzymatic hydrolysis and fermentation of banana pseudostem hydrolysate and demonstrated glucose release followed by fermentation using *Saccharomyces cerevisiae* [7].

Biological treatment of lignocellulosic biomass is attractive because microorganisms and their extracellular enzymes can modify the lignocellulosic structure and facilitate the release of fermentable carbohydrates [2,5]. *Trichoderma* species are particularly relevant because of their cellulolytic and hemicellulolytic enzyme systems and their established application in biological processing of lignocellulosic materials [5,7].

Following the release of fermentable sugars, *Saccharomyces cerevisiae* is widely employed for ethanol fermentation because of its strong fermentative capability and established tolerance to several fermentation conditions [8]. However, lignocellulosic hydrolysates may contain inhibitory compounds such as weak acids, furans and phenolic compounds, which can affect yeast growth and ethanol productivity [8,9].

Analytical characterization is therefore essential for evaluating biomass conversion. HPLC is widely used for the characterization and quantification of soluble sugars in biomass hydrolysates, while GC is commonly employed for volatile fermentation products. FTIR provides complementary information concerning the functional groups present in the resulting product [10–12].

The present study investigated the biological conversion of banana pseudostem followed by fermentation and characterization of the resulting product using HPLC, GC and FTIR spectroscopy. The analytical approach was designed to establish the presence of glucose in the treated biomass fraction and to obtain complementary evidence for ethanol in the fermentation-derived product.

2. MATERIALS AND METHODS

2.1 Banana pseudostem and biological treatment

Banana pseudostem was selected as the lignocellulosic agricultural residue for the study. The prepared biomass was subjected to biological treatment using *Trichoderma* according to the experimental protocol employed in the study.

Biological treatment was used to facilitate the breakdown or modification of the lignocellulosic matrix and increase the accessibility of fermentable carbohydrates. Biological pretreatment is recognized as one of the approaches investigated for improving lignocellulosic biomass conversion [2,5].

2.2 HPLC analysis

The treated banana pseudostem-derived hydrolysate was analyzed by HPLC to characterize the soluble carbohydrate fraction.

The chromatographic response was compared with the available glucose reference. A prominent peak corresponding to glucose was observed.

The result was interpreted qualitatively as evidence for the presence of glucose. Since a quantitative calibration curve was not available in the supplied analytical data, an absolute glucose concentration was not calculated.

2.3 Fermentation

The glucose-containing fraction obtained following biological treatment was subjected to fermentation using *Saccharomyces cerevisiae*. The use of *S. cerevisiae* for fermentation of lignocellulosic hydrolysates is well established in the literature [6–9].

2.4 GC analysis

Gas chromatography was performed for the ethanol reference standard and the fermentation-derived sample under the analytical conditions employed in the study.

The ethanol standard showed a prominent chromatographic peak. A major peak at a closely corresponding chromatographic position was observed in the fermentation-derived sample.

The sample peak was therefore interpreted by **direct comparison with the ethanol reference standard**.

This is an important point: the ethanol assignment is based on the correspondence between the sample chromatogram and the authentic standard, rather than on GC–MS library matching.

2.5 FTIR analysis

The fermentation-derived product was further characterized using FTIR spectroscopy.

The spectrum was examined for absorption features associated with hydroxyl, aliphatic C–H and C–O functional groups. FTIR spectroscopy has previously been demonstrated as a useful complementary approach for characterization of ethanol-containing fermented materials [11,12].

3. RESULTS AND DISCUSSION

3.*1 HPLC characterization*

The HPLC chromatogram of the biologically treated banana pseudostem fraction showed a prominent peak corresponding to glucose.

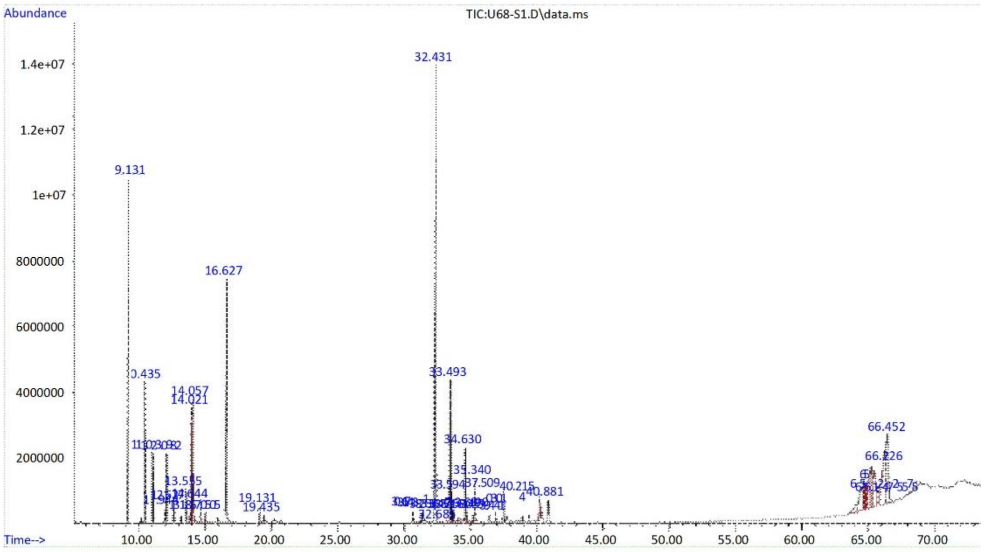

Figure 1. HPLC chromatogram of the glucose-containing fraction obtained following biological treatment of banana pseudostem.

The observation demonstrates that glucose was present in the treated biomass fraction and indicates the availability of a fermentable carbohydrate substrate. The finding is consistent with previous research showing that banana pseudostem can undergo biological or enzymatic treatment resulting in the release of fermentable sugars [5–7].

The presence of glucose is particularly significant because cellulose-derived glucose can serve as a substrate for subsequent ethanol fermentation by *S. cerevisiae* [7–9].

3.2 Fermentation

The glucose-containing fraction was subsequently subjected to yeast fermentation.

The conversion of fermentable sugars into ethanol by *S. cerevisiae* is well established. Previous studies on banana pseudostem have specifically demonstrated the feasibility of using treated BPS hydrolysates for ethanol fermentation [6,7].

Lignocellulosic hydrolysates can, however, contain compounds that negatively affect microbial fermentation. These include weak acids, furan derivatives and phenolic compounds generated or released during biomass processing [8,9]. The present study therefore focuses on analytical evidence for the resulting product rather than assuming complete conversion of the available carbohydrate.

3.3 GC characterization of ethanol

The GC chromatogram of the ethanol reference standard exhibited a prominent and well-defined peak. The fermentation-derived sample also exhibited a prominent peak at a chromatographic position closely corresponding to that of the ethanol standard.

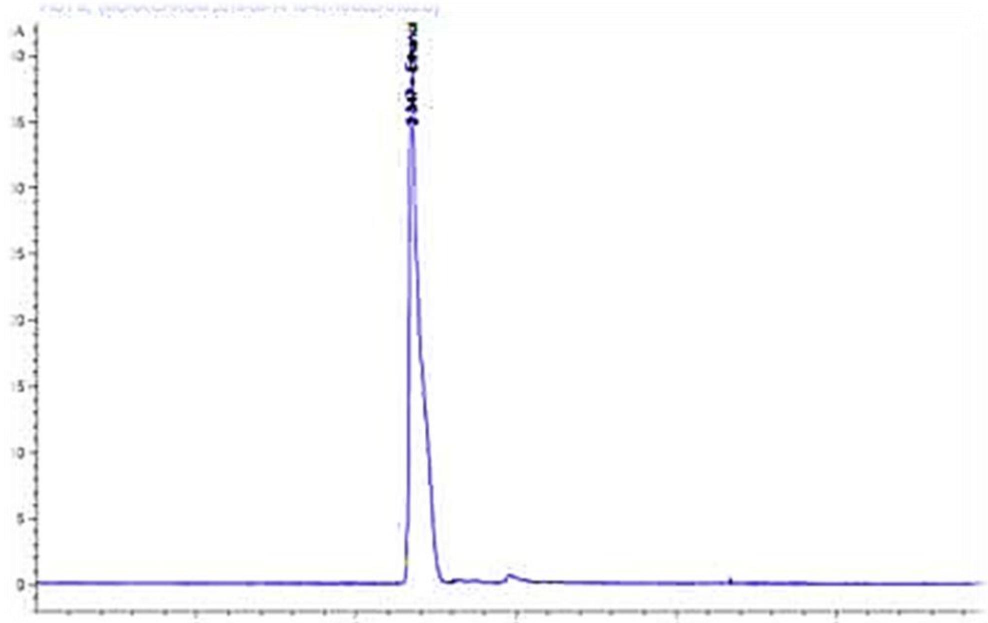

Figure 2. GC chromatogram of fermentation-derived banana pseudostem sample.

The close correspondence between the principal peak of the sample and the ethanol reference provides chromatographic evidence **consistent with the presence of ethanol** in the fermentation-derived product.

The use of an authentic reference standard is particularly important in chromatographic identification because retention behavior can depend on the chromatographic system and operating conditions.

Additional smaller peaks were observed in the sample chromatogram. These indicate that the recovered product contained other detectable volatile constituents. Therefore, the product should appropriately be described as an **ethanol-containing fermentation product**, rather than chemically pure ethanol.

Previous studies have demonstrated the feasibility of ethanol production from banana pseudostem hydrolysates [6,7], while broader studies of lignocellulosic fermentation have shown that hydrolysate composition can influence fermentation and the resulting product profile [8,9].

3.4 FTIR characterization

The FTIR spectrum of the fermentation-derived product exhibited a broad absorption in the approximately **3200–3500 cm⁻¹** region, consistent with O–H stretching vibrations.

Absorptions in the approximately **2850–3000 cm⁻¹** region were associated with aliphatic C–H stretching, while an absorption in the approximate **1000–1100 cm⁻¹** region was consistent with C–O stretching associated with alcohol-containing compounds.

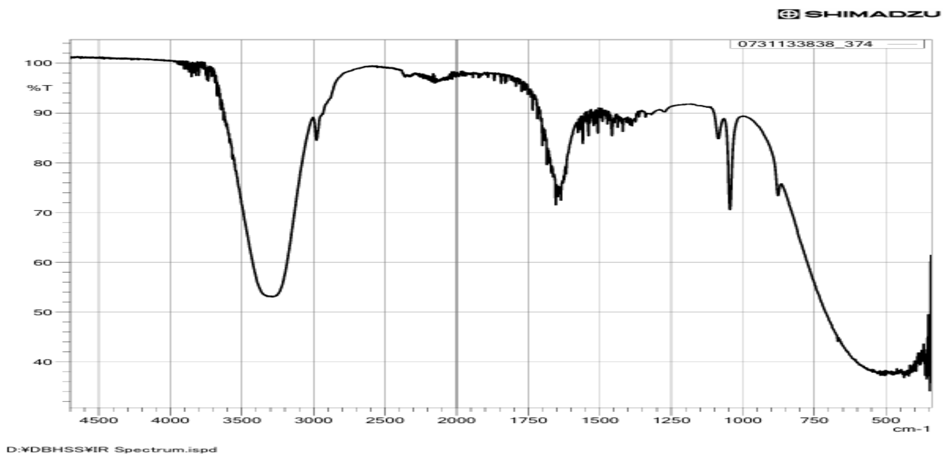

Figure 3. FTIR spectrum of the fermentation-derived product obtained from banana pseudostem.

The overall spectral pattern is compatible with an alcohol-containing product. FTIR has previously been employed for characterization and quantitative determination of ethanol in fermented alcoholic matrices [11,12].

The FTIR result should nevertheless be considered **supporting evidence rather than definitive identification by itself**, because functional-group absorptions can overlap among structurally related compounds.

3.5 Integrated interpretation

The analytical results provide complementary evidence for the proposed conversion pathway. The HPLC result establishes the presence of glucose in the treated fraction, while the GC result provides chromatographic evidence consistent with ethanol through comparison with an authentic reference. FTIR provides complementary evidence for alcohol-associated functional groups. Taken together, these independent observations support the proposed biological conversion of banana pseudostem-derived carbohydrate into an ethanol-containing fermentation product.

This interpretation is consistent with previous investigations demonstrating the potential of banana pseudostem for bioethanol production [5–7].

4. SIGNIFICANCE OF THE STUDY

Banana pseudostem represents an underutilized agricultural residue with potential for conversion into value-added products. Previous reviews have highlighted banana wastes, particularly pseudostem, as promising feedstocks for bioethanol production [5]. The conversion of lignocellulosic agricultural residues into fuels and chemicals is an important component of the broader biorefinery concept [3,4].

The present study combines three complementary analytical approaches:

- **HPLC** – evidence of glucose in the treated biomass;
- **GC** – chromatographic comparison with an ethanol reference standard; and
- **FTIR** – supporting evidence from alcohol-associated functional groups.

The use of multiple analytical techniques provides stronger evidence than relying on a single analytical measurement.

5. LIMITATIONS

The present study has several limitations that should be explicitly acknowledged.

First, the HPLC result demonstrates the presence of glucose but does not provide a validated glucose concentration because a quantitative calibration dataset was not available.

Second, the GC result provides evidence consistent with ethanol based on comparison with the ethanol standard, but it does not establish absolute sample purity.

Third, the FTIR spectrum provides complementary functional-group information but cannot independently establish ethanol identity and concentration in a complex sample.

Finally, the present analytical dataset does not permit calculation of:

- ethanol concentration;
- ethanol yield;
- fermentation efficiency;
- ethanol productivity;

- product purity; or
- overall carbon conversion.

Quantitative claims would require calibrated analytical measurements and appropriate experimental replicates.

6. CONCLUSION

The present study demonstrates the potential of banana pseudostem as a lignocellulosic agricultural residue for biological valorization.

The HPLC analysis demonstrated a prominent glucose-associated peak in the biologically treated biomass fraction. Following fermentation with *S. cerevisiae*, the resulting product exhibited a major GC peak corresponding closely to the ethanol reference standard. FTIR analysis further showed absorption features consistent with an alcohol-containing material.

The combined HPLC–GC–FTIR evidence therefore supports the biological conversion of banana pseudostem-derived carbohydrates into an **ethanol-containing fermentation product**. Previous studies independently support the feasibility of banana pseudostem conversion and fermentation for bioethanol production [5–7].

Importantly, the present results should not be interpreted as establishing absolute ethanol purity, concentration or fermentation yield. Such conclusions require quantitative and replicated analytical measurements.

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