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# PRODUCTION OF BIOETHANOL FROM BANANA WASTE INCLUDING ITS LEAF AND FRUIT PEEL

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## ABSTRACT

Banana peels are agricultural waste that has the capability to provide bio-ethanol as a renewable sort of energy. Pretreatment and reaction of lignocellulosic biomass are crucial steps in bio-ethanol production. Our study determined the potency of three pretreatment techniques specifically water, alkaline, and acidic pretreatments on the assembly of bio-ethanol. We tend to applied identical reaction technique mistreatment H2SO4 to any or all the pretreatment techniques. Bacteria was isolated from leaves and peels of banana .media optimization was done for the better production of bio-ethanol . Optimized media supplemented with pretreated banana waste for fermentation process. Downstream processing was carried out by distillation process.

**Key words**- Banana peels, lignocelluloses, bio-ethanol, distillation, media optimization

## INTRODUCTION

Fossil fuels burning at the recent would give to the environmental crisis widely **[1]**. The rise in demand of fossil fuels combined with depletion of this reserves oil has junction rectifier to the event of eco-friendly ideas **[2].** In addition, demand of the energy will increase with the rise of the planet population and urbanization **[3]** and therefore, development of bio-energy as energy may facilitate toscaleback these issu es.Bio-energy may be outlined as energy obtained from biomass, that is that the perishable fraction of merchandise, waste and residues from agriculture like vegetables and animal origin, biological science and connected industries and additionally, from the perishable fraction of business and municipal waste**[4].** Completely different kinds of bio-energy may be made from a large vary of biomass sources, as an example, agricultural residues **[5,6]**. There square measure several countries that use waste biomass as possibility instead of use food provide for energy production, like Southern Rhodesia and Australia. In Southern Rhodesia, some researchers are conducted on energy production from crop residues. The gross energy consumption wasabout a quarter mile in Southern Rhodesia that came from waste biomass **[7].** Meanwhile, banana waste hasbeen used to produce biogas using fed-batch digestion in Australia **[8].**In Australia, around half-hour of the harvested bananas ar rejected at the packing shed **[9].** Banana waste that are discarded thanks to the imperfections ar unremarkably drop as a large plenty of wastes, that ultimately cause contamination of water supply also as will have an effect on the atmosphere and health of living microorganisms **[10,20].** Thus, to avoid the environmental downside thanks to the decomposition of waste, it's usable to form energy from banana waste as biofuel production supply. so as to develop the new technologies and improve the on the market technologies relating to the biofuels production, it's essential to deal with the challenges and opportunities of befouls within the context of food security and property development wants **[11,17].**

## MATERIALS AND METHODOLOGY

### Collection of sample

For the following study, banana leaf and stem was collected from my village, Kanpur. Further the whole process done at MRD LifeSciences Pvt. Ltd. Lucknow.

### Isolation of bacteria

The samples were serially diluted in 0.85% NaCl solution by using serial dilution method and then spread over sterilized nutrient agar media and incubated at 37°C for 24 hours **[12, 13]**

### Purification of bacteria

The bacterial cultures were selected on the basis of different morphological parameters and then streaked in sterilized nutrient agar media by continuous quadrant streaking method. Then the cultures were incubated at 37°C for 24 hours.

### Screening and strain identification of bacteria

The cultures were screened on the basis of hydrolysis of starch content present in the banana and the estimation of enzyme is done by DNS test. Identification of the cultures was concluded by using Bergey's manual **[14].**

### Pretreatment of sample

The pretreatment of the sample was done by the acid base treatment of the banana leaf and peel **[15,16]**.

### Selection and optimization of Production media

Table 1:Media was optimized by one factor at a time method.

| Factors | Components | Concentration |
|---|---|---|
| Production Media | $KH_2PO_4$ | 6 g/l |
| | $NaH_2PO_4$ | 2 g/l |
| | Peptone | 5 g/l |
| | Dextrose | 8g/l |
| Carbon Sources | Dextrose | 8g/l |
| | Mannitol | 8g/l |
| | Maltose | 8g/l |
| | Fructose | 8g/l |
| Nitrogen Sources | Yeast | 5 g/l |
| | Peptone | 5 g/l |
| | $NH_4Cl$ | 5 g/l |
| | Malt extract | 5 g/l |

| | | |
|---|---|---|
| Metal Ion Sources | $FeSO_4$ | 0.2 g/l |
| | $MgsO_4$ | 0.2 g/l |
| | $CaCO_3$ | 0.2 g/l |
| | $ZnSO_4$ | 0.2 g/l |
| Salt Concentration | NaCl | 0.25 % |
| | NaCl | 0.5 % |
| | NaCl | 1 % |
| | NaCl | 1.5 % |

**Growth curve study**

The cultures were inoculated in sterilized fermentative media and the bacterial growth OD was taken after 1 hour of time interval **[18, 19]**.

**Fermentation**

The shake flask fermentation was performed for the production of bioethanol by supplementing 50% of pre-treated leaves and peels of banana in powder form. Incubated at 37ºC for 1 week.

**Distillation**

The supernatant was collected after centrifugation, boiled at 78ºC for collecting sample after condensation and then condensed sample was boiled at 65ºC. Further, the bioethanol testing was performed from the condensed product after double distillation.

**RESULTS**

**Sample collection**

a) Banana leaf b) Banana peel

Figure 1.Collected samples.

**Isolation and purification of bacteria**

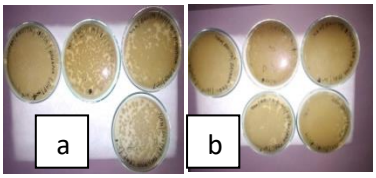


**Figure 2**- Isolation of bacteria from (a)Banana Leaves and (b)Banana Peels through the serial dilution and spread plat methods.

**Figure 3-** Streaking of isolated bacteria.

**Strain identification**

**Table 2**-Biochemical test for bacteria identification

| TEST NAME | LEAF BACTERIA | PEEL BACTERIA |
|---|---|---|
| Gram staining | Positive | Positive |
| Endospore test | Positive | Positive |
| Catalase test | Positive | Negative |
| Indole test | Negative | Negative |
| Urease test | Positive | Negative |
| Casein hydrolysis | Negative | Positive |
| Mannitol test | Negative | Negative |
| Amylase test | Negative | Negative |

**Screening**

**Table 3-** Screening of isolated bacteria for alcohol production.

| Culture | Result |
|---|---|
| 1 (Leaf bacteria) | ++ |
| 2 (Leaf bacteria) | + |
| 3 (Peel bacteria) | +++ |
| 4 (Peel bacteria) | ++ |

**Media selection and optimization**

**Table 4-** OD of optimized media at 620 nm and 540 nm.

| Factors | Component | OD at 620 nm | |
|---|---|---|---|
| | | Leaf isolate | Peel isolate |
| Production media | | 0.13 | 0.17 |
| Carbon Sources | Dextrose | 0.13 | 0.13 |
| | Mannitol | 0.20 | 0.20 |
| | Maltose | 0.33 | 0.16 |
| | Fructose | 0.15 | 0.30 |
| Nitrogen Sources | Yeast | 0.11 | 0.18 |
| | Peptone | 0.21 | 0.20 |
| | NH4Cl | 0.20 | 0.11 |
| | Malt extract | 0.15 | 0.23 |

| | | | |
|---|---|---|---|
| Metal ion Sources | FeSO4 | 0.46 | 0.44 |
| | MgSO4 | 0.22 | 0.54 |
| | CaCO3 | 0.46 | 0.44 |
| | ZnSO4 | 0.53 | 0.48 |
| Salt concentration | 0.25%NaCl | 0.31 | 0.32 |
| | 0.5%NaCl | 0.74 | 0.84 |
| | 1%NaCl | 0.89 | 0.92 |
| | 1.5%NaCl | 0.32 | 0.52 |

| Factors | Component | OD at 540 nm | |
|---|---|---|---|
| | | Leaf isolate | Peel isolate |
| Production media | | 0.56 | 0.61 |
| Carbon Sources | Dextrose | 1.36 | 1.43 |
| | Mannitol | 1.37 | 1.20 |
| | Maltose | 0.35 | 0.45 |
| | Fructose | 1.03 | 1.62 |
| Nitrogen Sources | Yeast | 1.11 | 1.38 |
| | Peptone | .98 | 1.00 |
| | NH4Cl | 1.02 | 0.85 |
| | Malt extract | 1.03 | 1.14 |

| | | | |
|---|---|---|---|
| Metal ion Sources | FeSO4 | 0.83 | 0.84 |
| | MgSO4 | 0.84 | 0.90 |
| | CaCO3 | 0.86 | 1.00 |
| | ZnSO4 | 0.93 | 1.14 |
| Salt concentration | 0.25%NaCl | 1.48 | 1.52 |
| | 0.5%NaCl | 1.51 | 2.62 |
| | 1%NaCl | 1.59 | 2.79 |
| | 1.5%NaCl | 1.53 | 1.63 |

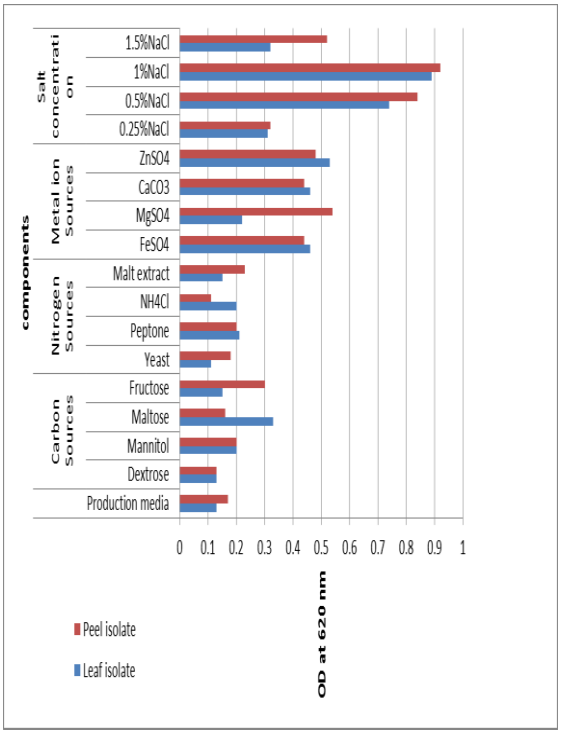


**Figure 4-** optimized media by one factor at a time.

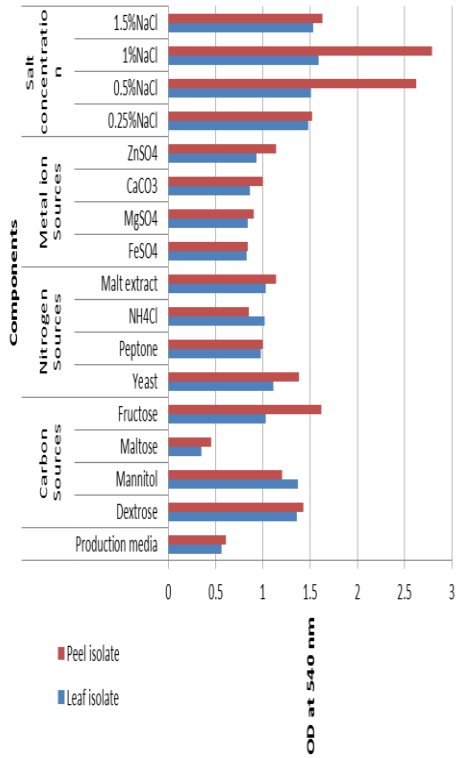


**Figure 5-** Optimized media for C, N metal ions and salts.

**Estimation**

Table 6- Estimation of alcohol after down streaming.

| S.NO. | Sample | OD 540 nm |
|---|---|---|
| **1** | Blank | 0 |
| **2** | Control | 1.34 |
| **3** | Distilled product | 1 |
| **4** | Double distilled | 0.4 |

## DISCUSSION

Isolated bacteria is important microorganism can be used to produce ethanol from a various substrate present in banana waste , it produces ethanol in large quantity and has the advantage over other organism of resisting multiple inhibitors such as furanse, phenolic components and organic acid. The results obtained from the glucose analysis showed a great promise of producing bio-ethanol from all the pretreatment techniques employed .the result of ethanol analysis is presented in graph no . optical density obtained from alcohol test confirmed the presence of bio-ethanol in all the pretreatment technique

## CONCLUSION

The study investigated the feasibility of producing from banana peels and leaves as source of lignocelluloses biomass. Three different pretreatment techniques were employed as the first step in the experimental design the techniques produced the different concentration of reducing sugars after hydrolysis with sulfuric acid.

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