Bio-ethanol Fuel Production from Rotten Banana Waste

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Abstract: Banana is widely available resource of lignocellulosic mass. It is second most cultivated fruit in world. During harvesting process, for production of 100 kg of banana there is generation of 4 tones of waste that is four times of waste are generated. The waste has been dumped in soil which can affect the environment and health of living organism. In present paper is study of generation of bio-ethanol from rotten banana waste by using enzymatic hydrolysis and fermentation. In this method we are using saccharomyces cerevisiae type II as yeast and two yeasts pectinase and cellulase. The study shows different factors affect the yield of bio-ethanol production.

Keywords: Enzymatic hydrolysis, fermentation, saccharomyces cerevisiae, cellulase, pectinase

1. Introduction

The unsustainable energy resources like fossil fuel are going to be depleted in the next few decades and the trends in bio-fuel production from biomass are gaining popularity to encounter the expected energy crisis in the world. Ethanol is the most widely used liquid bio-fuel and is produced as a result of fermentation process from sugars, starches or cellululosic biomass including fruit wastes. In this study rotten banana is utilized for bio-ethanol production. Banana waste is used to produce bio-ethanol by fermentation using yeast Saccharomyces Cerevisiae. The ethanol production from lignocelluloses material is oxygenated and sulphur free. Carbon in ethanol has a vegetable origin and when it goes on combustion process, it will not contribute to produce CO₂ Emission. The renewable ethanol content, which is produced from banana waste is helpful in a net reduction in the emission of CO₂, CO and hydrocarbon. Ethanol itself burns cleaner and burns more completely than gasoline so it can be blended with petrol.

Banana is the second most cultivated fruit in the world. In Malaysia, banana plant covers 30000 hectares with a total production of 540000 metric tons per year. For production of 100 kg of banana there was generation of 4 tons of waste that is four times of waste were generated. Ethanol itself burns cleaner and burns more completely than gasoline so it is blended with petrol.

Basicall plant material mainly composed of three units namely cellulose, Hemicelluloses and lignin. Cellulose is about 40-50 % and it is main component of plant cell likewise hemicelluloses is about 20-30%. Hemicellulose nothing but macromolecules of repeated polymers of pentose and hexoses. Lignin is about 10-25% and it contains series of three aromatic alcohols coniferyl alcohol, p-coumaryl alcohol and sinapyl alcohol produced from biosynthetic process and forms a protective seal around the other two components cellulose and hemicellulose.

The process of converting lignocelluloses biomass to bio-ethanol requires five distinct steps:-

1) Biomass delignification to release ocellulose and hemicellulose from lignin–cellulose-hemicellulose complex
2) Depolymerisation of hydrolysis of the cellulose and hemicelluloses to produce free sugars
3) Fermentation of the free sugars to produce ethanol
4) Ethanol recovery
5) Effluent Treatment

In present paper work we plan to investigate the different parameters like temperature, shaking hours and water content on bio-ethanol production by using rotten banana mixture along with proper concentration of enzyme and yeast during fermentation. And to find out is it suitable for use in engine as a fuel by checking properties like viscosity, Pₕₒ and metal content.

2. Experimentation

2.1 Materials

The banana waste bring from local market from Pune, India. The enzymes used pectinase and cellulase. These enzymes brought from supplier data global trading, Ahmedabad. The enzymes used 0.3 units per mg. Yeast for this fermentation process was Saccharomyces cerevisiae type II. It is activated by hot water bath at 37°C and then dried fast to yield 90% active, viable yeast in sigma solid form.

2.2 Fermentation procedure

Part 1

Take four sets, 1200 gm of rotten banana was taken and it was washed with distilled water. It was cut by using knife and mash wash prepared by using automatic juice blender. Take 12 sterile bottles each contain 100 gm of mash.

SET1

The banana mash then taken into 3 sterile bottles add 25 ml water in each bottle. Add 3 gm/lit of yeast S.cerevisiae in each bottles closely tightened so that environmental related to anaerobic condition been provide and it was placed in incubator at 23°C, 30°C and 35°C, each at particular temperatures for 3 days of fermentation.
Viscosity was determined by refractometer in % brix unit, before and after fermentation. Total soluble solids content was detected by digital meter with the help of P2.3 to total soluble solids media. The mother liquor analysis was carried with Part 1 and Part 2 samples were only days of fermentation changes to 1.3 and 5 days.

Third set of three bottles of mash then added with 0.4 ml of cellulases alone and it is heated to 65°C for 2 hrs and it is cooled to room temperature and used in fermentation process for 3 days with S. cerevisiae as yeast as per set 1.

Fourth set of three bottles of mash in which we did all the activity as in set 1,2 and 3 that is in fourth set we use both pectinases and cellulases as enzyme and S. cerevisiae as yeast. The fermentation process was carried for 3 days at different temperature 23°C, 30°C and 35°C.

Part 2
Take three sets, gm of rotten banana was taken and it was washed with distilled water. It was cut by using knife and mash wash prepared by using automatic juice blender. Take 12 sterile bottles each contain 100 gm of mash.

First set of three bottles taken in which we did the activity as per part 1 but only shaking hrs changing to 0,3 and 6 hrs. And fermentation process was carried out at 35°C.

Second set of three bottles taken in which same process was carried out shaking 6 hrs, fermentation temperature 35°C only change in water content in each bottles was 15%, 25% and 35% respectively.

Third set of three bottles taken same amount of enzyme, yeast used. Shaking 6 hrs, fermentation temperature 35°C only days of fermentation changes to 1.3 and 5 days. Then Part 1 and Part 2 samples were filtered with the help of filter media. The mother liquor analysis was carried with respect to total soluble solids, pH, glucose analysis, bio-ethanol concentration, viscosity.

2.3 Analytical Analysis

pH
pH was checked before and after the fermentation process with the help of pH meter.

Total soluble solids
Total soluble solids content was detected by digital refractometer in % brix unit, before and after fermentation.

Viscosity
Viscosity was determined by viscometer before and after fermentation.

Elemental Analysis
Metal contents P, Ca, Mg, Fe, Pb, Cu, etc were detected by using multi-element spectrometry.

Bio-ethanol concentration
Bio-ethanol concentration was detected by potassium dichromate reagent solution, S-diphenyl carbazide solution and 40% potassium sodium tartrate solution. This method was of williams and Darwin (1950).

The potassium dichromate reagent solution prepared by dissolving 1 gm of potassium dichromate dissolved in 100 ml 6N sulphuric acid. The prepared solution was properly shake in order to prepare homogeneous solution.

The S-diphenyl carbazide solution prepared by 1 gm of S-diphenyl carbazide dissolved in 1 ml 95% ethanol solution and supertant is collected. A homogenous mixture of 1ml of potassium dichromate, 1 ml of S-diphenyl carbazide and 1ml of mash sample taken in test tube. The test tube was covered by paraffin film. The test tube heat to 90°C by using hot water bath for 5 to 15 minute until look like red brown colour then add 1ml of potassium sodium tartarate of 40% strength to stabilize the colour of solution. Then solution is cooled to room temperature in cold water bath. The Ethanol absorbance values measured at 575 nm wavelength by spectrometer.

Glucose Estimation
Glucose estimated in samples detected by DNS solution that dinitrosalicyclic acid reagent solution, it is produced as 10 gm of DNS acid, 2 gm of phenol, 0.5 gm of sodium sulphite, 10 gm of NaOH, and mixed well in 1 lit of water. Out of this solution, 0.3 ml of DNS reagent added in 3ml of sample in lightly capped test tube. Then it was heated to 90°C until red brown colour appeared. Then add 1ml of 40% potassium tartarate solution to stable the colour. Then it was cooled to room temperature in cooled water bath. The absorbance value of the reducing sugar measured using spectrophotometer at 575 nm.

3. Results and Discussion

3.1 Effect of Temperature

The effect of temperature on bio-ethanol concentration was tabulated below:

<table>
<thead>
<tr>
<th>Temperature</th>
<th>Bio-Ethanol concentration%(V/V)</th>
</tr>
</thead>
<tbody>
<tr>
<td>23°C</td>
<td>5.39</td>
</tr>
<tr>
<td>30°C</td>
<td>5.88</td>
</tr>
<tr>
<td>35°C</td>
<td>6.21</td>
</tr>
</tbody>
</table>

As from the analysis. The Bio-ethanol Concentration was increased with temperature. The highest concentration 6.21%(v/v) of bio-ethanol at 35°C during fermentation process.

As we plot the graph of Temperature Vs Bio-ethanol concentration.

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The effect of temperature of total soluble solids and pH on banana mash was tabulated as before and after fermentation as follows:

<table>
<thead>
<tr>
<th>Temperature</th>
<th>Total soluble solids in Brix</th>
<th>pH</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Before</td>
<td>After</td>
</tr>
<tr>
<td>23°C</td>
<td>17.20</td>
<td>15.47</td>
</tr>
<tr>
<td>30°C</td>
<td>17.33</td>
<td>13.67</td>
</tr>
<tr>
<td>35°C</td>
<td>17.0</td>
<td>11.53</td>
</tr>
</tbody>
</table>

3.2 Effect of Shaking Hrs

The concentration of bio-ethanol at different shaking hrs is tabulated. The fermented banana mash without shaking only produced 5.86% followed by 3 hrs shaking period 6.35% and 6 hrs produce 6.55% (v/v) ethanol.

<table>
<thead>
<tr>
<th>Shaking Hrs</th>
<th>Bioethanol Concentration</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>5.86</td>
</tr>
<tr>
<td>3</td>
<td>6.35</td>
</tr>
<tr>
<td>6</td>
<td>6.55</td>
</tr>
</tbody>
</table>

We plot above table as follows:

The Effect of shaking hrs on the soluble solid and pH of banana mash tabulated below before and after fermentation:

<table>
<thead>
<tr>
<th>Shaking hrs</th>
<th>Total soluble solids</th>
<th>pH</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Before</td>
<td>After</td>
</tr>
<tr>
<td>0</td>
<td>17</td>
<td>10</td>
</tr>
<tr>
<td>3</td>
<td>17</td>
<td>10.47</td>
</tr>
<tr>
<td>6</td>
<td>17</td>
<td>10.07</td>
</tr>
</tbody>
</table>

3.3 Effect of water content

The lowest volume of 4.30% (v/v) ethanol was produced from the fermentation process without water. The highest volume of ethanol produced 6.38% (v/v) produced with 35% of water.

<table>
<thead>
<tr>
<th>% of water contain</th>
<th>Bio-ethanol concentration</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>4.33</td>
</tr>
<tr>
<td>15</td>
<td>5.37</td>
</tr>
<tr>
<td>25</td>
<td>5.85</td>
</tr>
<tr>
<td>35</td>
<td>6.38</td>
</tr>
</tbody>
</table>

The values of total soluble solids for fermented banana mash lower than before fermentation. The banana mash after fermentation without water found higher value of soluble total soluble solids followed by 15,35 and 25% of water. From pH measurement, pH of fermented banana of water content 35% had the highest pH compared to others.

3.4 Glucose Analysis

For DNS method the glucose Concentration for different Fermentation period tabulated below:

<table>
<thead>
<tr>
<th>Time (Days)</th>
<th>Glucose concentration % (v/v)</th>
<th>Biethanol concentration % (v/v)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>13</td>
<td>0</td>
</tr>
<tr>
<td>1</td>
<td>3.7</td>
<td>5.6</td>
</tr>
<tr>
<td>3</td>
<td>3.32</td>
<td>5.85</td>
</tr>
<tr>
<td>5</td>
<td>0.6</td>
<td>6.09</td>
</tr>
</tbody>
</table>

From 3 days to 5 days, there was slight increase in ethanol from 5.85 to 6.09% (v/v). The large amount of glucose used at the initial stage caused to produce 5.6% (v/v) of ethanol production. The highest amount of glucose shows lowest amount of glucose concentration 0.6% (v/v).

3.5 Effect of different enzyme

The effect of different enzyme treatment on bio-ethanol treatment on bio-ethanol concentration is tabulated below:

<table>
<thead>
<tr>
<th>Enzyme</th>
<th>Bio-ethanol concentration % (v/v)</th>
</tr>
</thead>
<tbody>
<tr>
<td>No enzyme</td>
<td>5.80</td>
</tr>
<tr>
<td>Cellulase</td>
<td>6.65</td>
</tr>
<tr>
<td>Pectinase</td>
<td>7.02</td>
</tr>
<tr>
<td>Pectinase + cellulase</td>
<td>7.06</td>
</tr>
</tbody>
</table>

The combination S. cerevisiae + pectinase + cellulase gives highest bio-ethanol concentration.
4. Conclusions

It was concluded that banana fruit waste could be used to produce good quality of bio-ethanol, which can be used in engine of motor vehicles. The engine produced low emissions CO and CO₂; it can be a recycling process for solid waste management.

References


