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Production of Bioethanol from Sugar Syrup

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Abstract:
Ethanol production from pure sugar cane syrup (SCS) were carried out to compare the efficiency of batch fermentation with commercial glucose using Saccharomyces cerevisiae baker's yeast and Saccharomyces cerevisiae CSI-1 (JCM 15097). Fermentation of Baker's yeast and CSI-1 in SCS medium supplemented with 0.5% (w/v) of yeast extract at 37oC, initial pH adjusted to 5.5, agitation 200 rpm, produced high concentration of ethanol in 1.5L lab scale fermentor. Ethanol production by dry Bakers's reached 138g/L and yield 156.9% of theoretical yield. For CSI-1, a maximal concentration of 73.3g/L and a yield of 88.9% of theoretical yield were obtained from fresh SCS. The result from this research demonstrate that in Malaysia, sugar cane syrup could be employed as an alternative renewable carbon source for ethanol production using batch fermentation with Saccaharomyces cerivisiae.

Key Words: Ethanol, fermentation, Saccharomyces cerevisiae, sugar cane syrup

1.INTRODUCTION

Several renewable substrates are under investigation as feedstock for bioconversion to fuel ethanol. In temperate climates, grain starch is the basic raw material for fermentation ethanol due to its avaibility and low cost. In tropical country such as Malaysia, sugar cane juice can be the sources of sugar for fermentation into ethanol. Sugar cane is a highly productive land plant that uses the C4 pathway of photosynthesis, which confers higher potential light, water and nitrogen use efficiencies, than the alternative C3 pathway (De Souza & Buckeridge, 2010). In general, the ethanol fermentation of these saccharine materials is much simpler than the fermentation of grain starch (Hodge and Hildebrand, 1954; Maiorella, 1985). Unlike grain starch, in which starch has to be broken down into sugars with expensive enzymes before it can be fermented, the entire sugar cane stalk is already contain high sugar and it starts to ferment almost as soon as it's cut . Currently in Brazil, sugar cane worts of 16%w/v to 20% w/v are routinely fermented to produce 7.5 to 10.0 % (w/v) ethanol (Laluce, 1991). This project studied the use of sugar cane syrup for ethanol production by Mauripan dried instant yeast and Saccharomyces cerevisiae CSI-1 (JCM 15097) using a nonaerated bench-top fermentor. Comparative studies were performed between 2 strains of Saccharomyces cerevisiae grown on 15% to 19% (w/v) of either commercial glucose (CG) or sugar cane syrup (SCS) with the initial pH set at 5.5 and uncontrolled throughout the experiment. The objectives of this study were to quantify the production of ethanol from sugar cane syrup under batch fermentation using Baker's yeast and CSI-1 and to determine the feasibility of planting sugar cane locally to reduce import of the raw material.

2. Body of Paper

Bioethanol is an energy saving fuel which is readily available for combustion engines made from biomass. Production of bioethanol lowers both consumption of crude oil and greenhouse gas emissions from transport and traffic because it only releases the same amount of carbon dioxide as plants bound while growing. Because of its high octane number, low octane number and high heat of vaporization impede self-ignition in the diesel engine; it is suitable for the mixed fuel in the gasoline engine. Therefore, ignition improver, glow-plug, surface ignition, and pilot injection are applied to promote self-ignition by using diesel-bioethanol-blended fuel. Disadvantages of bioethanol include its lower energy density than gasoline, its corrosiveness, low flame luminosity, lower vapor pressure (making cold starts difficult), miscibility with water, and toxicity to ecosystems. Bioethanol is produced by a biological fermentation process in which organic material is converted by microorganisms to simpler compounds, such as sugars. These fermentable compounds are then fermented by microorganisms to produce ethanol and CO2. Besides, enzyme also produced by the microorganisms to catalyze chemical reactions that hydrolyze the complicate substrates into simpler compounds for carbon source. Several strains of microorganisms, bacteria, yeasts, and fungi have been reportedly used for the production of ethanol

Both yeasts (Saccharomyces cerevisiae, S. uvarum S. carlsbergensis, Candida brassicae, C. utilis, Kluyveromyces fragilis, K. lactis) and bacteria (Zymomonas mobilis) have been employed for ethanol production in industries. • The commercial production is carried out with Saccharomyces cerevisiae. On the other hand, S. uvarum has also largely been used. The Candida utilis is used for the fermentation of waste sulphite liquor since it also ferments pentoses. • Recently, experimentation with Schizosaccharomyces has shown promising results. When whey from milk is used, strain of K. fragilis is recommended for the production of ethanol. It is also found that Fusarium, Bacillus and Pachysolen tannophilus (yeast) can transform pentose sugars to ethanol. • It is noteworthy that the ethanol at high concentration inhibits the yeast. Hence, the concentration of ethanol reduces the yeast growth rate which affect the biosynthesis of ethanol. • The bacteria Zymomonas mobilis has a merit over yeast that it has osmotic tolerance to higher sugar concentration. It is relatively having high tolerance to ethanol and have more specific growth rate.

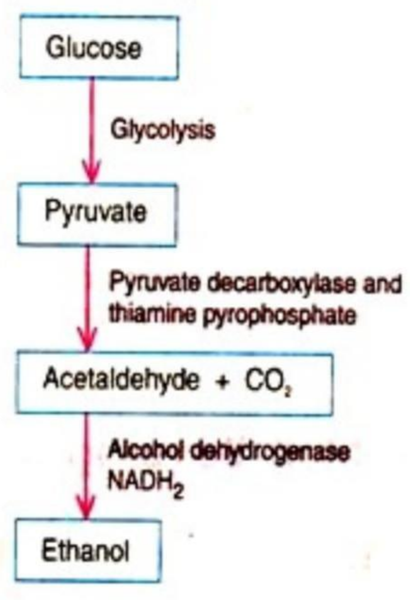

Fig-01

Procedure

1. Procuring the grain or plant

2. Converting this to sugar

3. Fermentation

4. Distillation

• On industrial scale, ethanol is produced by the fermentation of molasses. Molasses is the mother liquor left after the crystallization of sugarcane juice. It is a dark colored viscous liquid. Molasses contains about 60% fermentable sugar

1) Ammonium sulphate.

If nitrogen content of molasses is less, it is fortified with ammonium sulphate to provide adequate supply of nitrogen to yeast.

2) Addition of sulphuric acid

Fortified solution of molasses is then acidifies with small quantity of sulphuric acid. Addition of acid favours the growth of yeast but unfavours the growth of useless bacteria

3) Fermentation

The resulting solution is received in a large tank and yeast is added to it at 35°C and kept for 2 to 3 days. During this period, enzymes sucrose and zymase which are present in yeast, convert sugar into ethyl alcohol

$$C_{12}H_{22}O_{11} + H_2O \rightarrow C_6H_{12}O_6 + C_6H_{12}O_6$$

$$C_6H1_2O_6 \rightarrow C_2H_5OH + 2CO_2$$

4) Fractional distillation

Alcohol obtained by the fermentation is called "wash" which is about 15% to 18% pure. By using fractional distillation technique, it is converted into 92% pure alcohol which is known as rectified spirit or commercial alcohol. Production of ethanol from various feed stocks involves the following steps. 1) Feed preparation

2) fermentation
3) distillation
4) dehydration and
5) denaturing. Alcohol (Ethanol) Production

i) Feed preparation

The first step in making ethanol is to prepare the feedstock to enter the fermentation process. Cereal grains, such as corn, rye, rice, barley, soybeans, wheat, and plants like sugar cane are the major sources of feedstock's of fermentation. Some producers use high starch plants such as potatoes. Many different methods are used to prepare the feedstock to enter the fermentation process. All of the different processes ultimately produce a liquid solution that contains fermentable sugars. These solutions are clarified and heated to high temperature for 20 to 30 minutes to reduce the bacterial levels which can harm the performance of the process. After this treatment the liquid mixture is removed and subjected to fermentation process. If sugar cane is used as a feedstock, the liquid mixture is said to be sugarcane juice or molasses.

ii) Fermentation of sugars

The liquid mixture obtained in the above process is subjected to fermentation process by adding yeast cells. Zymase, an enzyme from yeast, changes the simple sugars into ethanol and carbon dioxide. The enzymatic reaction carried over by the yeast in fermentation produces mainly ethanol, CO2 and heat. The fermentation reaction is actually very complex and the resulting product is similar to beer or wine. The impure culture of yeast Produces varying amounts of other substances, including glycerin, methanol and various organic acids. After fermentation, the liquid is subjected to distillation to separate alcohol from water.

iii) Distillation

Ethanol produced by fermentation ranges in concentration from a few percent up to about 14 percent balance is being water and other components. The boiling point of ethanol (78.4'C) is slightly lower than the boiling point of water (100°C). Since the difference in the boiling point of these materials is low they cannot be completely separated by distillation. Instead, an azeotropic mixture (i.e. a mixture of 96%ethanol and 4% water) is obtained. Azeotropic mixture of alcohol cannot be further concentrated by distillation. Distillation is used to produce Rectified Spirit (RS, 94%v/v ethanol). 2) Ammonium sulphate. If nitrogen content of molasses is less, it is fortified with ammonium sulphate to provide adequate supply of nitrogen to yeast. • 3) Addition of sulphuric acid Fortified solution of molasses is then acidifies with small quantity of sulphuric acid. Addition of acid favours the growth of yeast but unfavours the growth of useless bacteria • 4) Fermentation The resulting solution is received in a large tank and yeast is added to it at 35°C and kept for 2 to 3 days. During this period, enzymes sucrose and zymase which are present in yeast, convert sugar into ethyl alcohol $C_{12}H_{22}O_{11}$ + H_2O à $C_6H_{12}O_6$ + $C_6H_{12}O_6$ $C_6H_{12}O_6$ à C_2H_5OH + $2CO_2$ 5. Fractional distillation Alcohol obtained by the fermentation is called "wash" which is about 15% to 18% pure. By using fractional distillation technique, it is converted into 92% pure alcohol which is known as rectified spirit or commercial alcohol. Production of ethanol from various feed stocks involves the following steps. I) Feed preparation 2) fermentation 3) distillation 4) dehydration and 5) denaturing. Alcohol (Ethanol) Production i) Feed preparation The first step in making ethanol is to prepare the feedstock to enter the fermentation process. Cereal grains, such as corn, rye, rice, barley, soybeans, wheat, and plants like sugar cane are the major sources of feedstock's of fermentation. Some producers use high starch plants such as potatoes. Many different methods are used to prepare the feedstock to enter the fermentation process. All of the different processes ultimately produce a liquid solution that contains fermentable sugars. These solutions are clarified and heated to high temperature for 20 to 30 minutes to reduce the bacterial levels which can harm the performance of the process. After this treatment the liquid mixture is removed and subjected to fermentation process. If sugar cane is used as a feedstock, the liquid mixture is said to be sugarcane juice or molasses. ii) Fermentation of sugars The liquid mixture obtained in the above process is subjected to fermentation process by adding yeast cells. Zymase, an enzyme from yeast, changes the simple sugars into ethanol and carbon dioxide. The enzymatic reaction carried over by the yeast in fermentation produces mainly ethanol, CO_2 and heat. The fermentation reaction is actually very complex and the resulting product is similar to beer or wine. The impure culture of yeast Produces varying amounts of other substances, including glycerin, methanol and various organic acids. After fermentation, the liquid is subjected to distillation to separate alcohol from water. iii) Distillation Ethanol produced by fermentation ranges in concentration from a few percent up to about 14 percent balance is being water and other components. The boiling point of ethanol (78.4'C) is slightly lower than the boiling point of water (100°C). Since the difference in the boiling point of these materials is low they cannot be completely separated by distillation. Instead, an azeotropic mixture (i.e. a mixture of 96%ethanol and 4% water) is obtained. Azeotropic mixture of alcohol cannot be further concentrated by distillation. Distillation is used to produce Rectified Spirit (RS, 94%v/v ethanol). iv) Dehydration of Alcohol Pure alcohol can? be obtained from distillation since it forms azeotrope with water at 96% (vlv). Ethanol or absolute alcohol is produced by dehydration of rectified spirit. Commercially available technologies for dehydration of rectified spirit are a) Azeotropic distillation and b) Molecular Sieve Technology.

iv) Dehydration of Alcohol

Pure alcohol can? be obtained from distillation since it forms azeotrope with water at 96% (vlv). Ethanol or absolute alcohol is produced by dehydration of rectified spirit. Commercially available technologies for dehydration of rectified spirit are a) Azeotropic distillation and b) Molecular Sieve Technology.

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Process Diagram

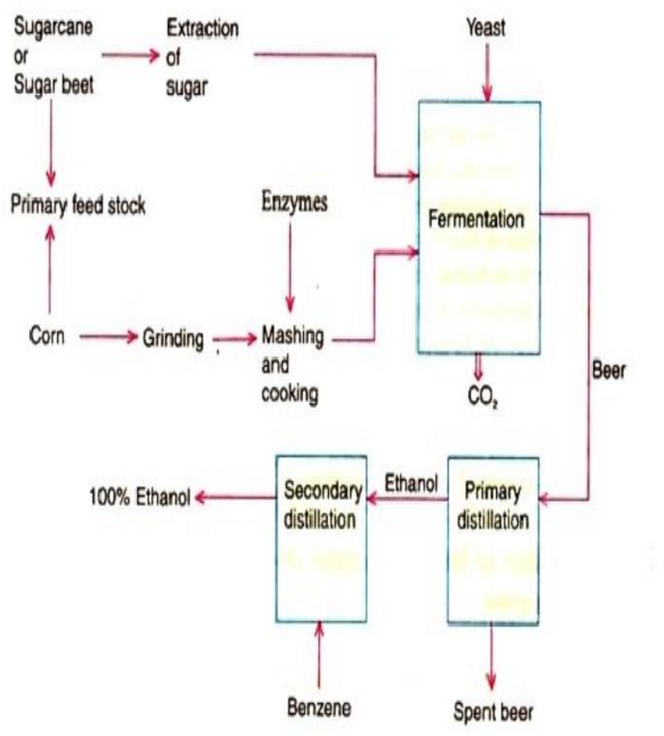

Fig-2

Fermentation:

• Ethanol is produced by continuous fermentation. Hence, large fermenters are used for continuous manufacturing of ethanol. The process varies from one country to another. India, Brazil, Germany, Denmark have their own technology for ethanol production.

• The fermentation conditions are almost similar (pH 5, temperature 35°C) but the cultures and culture conditions are different. The fermentation is normally carried out for several days but within 12h starts production.

• After the fermentation is over, the cells are separated to get biomass of yeast cells which are used as single cell protein (SCP) for animal's feed. The culture medium or supernatant is processed for recovery of ethanol

. • Ethanol is also produced by batch fermentation as no significant difference is found both in batch and continuous fermentation.

• Although as stated earlier within 12h Saccharomyces cerevisiae starts producing ethanol at the rate of 10% (v/v) with 10-20g cells dry weight/lit. The reduction in fermentation time is accomplished use of cell recycling continuously in fermentation.

3. CONCLUSIONS

The successful operation of ethanol fermentation depends on the viability of the yeast strains used with regard to a number of stress factors occurring during the process. Thus, the
selection process is important for effi cient ethanol fermentation. In addition to ethanol production from cane molasses by K. marxianus DMKU3- 1042 (Limtong et al., 2007), the results from this research demonstrated that sugar cane syrup could be employed as an alternative renewable carbon source for ethanol production using this effi cient thermo-tolerant yeast strain as well. K. marxianus can use sucrose as a carbon source as the inulinase (an enzyme present in the yeast

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