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Review

Bioethanol sources in Pakistan: A renewable energy resource

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Demand of bioethanol is increasing rapidly as fuel to replace the use of gasoline due to the recent debates on environmental issues and in the increase of gas prices. To combat these threats, bioethanol and biodiesel are being produced as alternative fuel sources. A wide range of substances can be used for bioethanol production. Biofuel in form of bioethanol can be produced using agricultural wastes by the use of enzymes, and hence the main objective of this approach is to get benefit of the agricultural wastes. The basic steps included in ethanol production from agricultural wastes composed of milling/grinding, cooking, saccharification, fermentation, purification/distillation and the last step is hydration. In every step, there are various strategies to increase bioethanol production and its usefulness. Bioethanol fuel obtained from starchy and lignocelluloses agricultural biomass is being gradually improved to minimize the use of petroleum due to decrease in the petroleum resources, and to diminish the effect of petrol on the environment.

Key words: Bioethanol, alternative fuel sources, fermentation.

INTRODUCTION

The oil reservoirs are diminishing rapidly and as a result gases from the greenhouse are released daily. Global warming significantly increases due to the use of fossil fuels in the form of oil, natural gas and coal, on which modern society depends for energy. Fuel ethanol which is produced from renewable sources can be utilised as an alternative to these fuels. This ethanol fuel is environmentally friendly and helps in saving national energy, as well in improvement of economy (Bin and Yanpin, 2007). Scientists are trying to protect the environment and to find alternative means of fossil fuels through biological ways and hence much research has been put into producing biofuels. Agricultural wastes materials are inexpensively found outside the food chain of human in large amount and can be obtained through out the season. These agricultural biomasses are the potential feedstock for bioethanol production, including the cellulosic biomass, as well as starchy waste agricultural materials. It is very important for humans to generate bioenergy and other bio-based products from these natural biological renewable resources (Percival et al., 2006) since agricultural and various industries generate huge volumes of starchy and cellulosic agricultural waste materials. Several bio-products can be produced from these agricultural biomasses, which have vast potential to be utilized for this purpose (Das and Singh, 2004).

To reduce the emission of gases from greenhouse by the production of biofuel and biochemical, these agricultural waste-based substances provide low-cost and uniquely sustainable resources, improving the security of energy, developing the economy, as well as cleaning the environment and atmosphere by the disposing of problematic solid wastes and getting wealth out of wastes. Many scientists have achieved improvements in bioethanol production from agricultural waste material. Bioethanol is made by the yeast *Saccharomyces cerevisiae* from sugars obtained from agricultural wastes.

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Table 1. Annual ethanol fuel production by country (2007 to 2010) in top 10 countries/regional blocks (millions of U.S. liquid gallons per year).

Country	2010	2009	2008	2007
United States	13,230.00	10,600.00	9,000.00	6,498.60
Brazil	6,921.54	6,577.89	6,472.2	5,019.2
European Union	1,176.88	1,039.52	733.60	570.30
China	541.55	541.55	501.90	486.00
Thailand	-	435.20	89.80	79.20
Canada	356.63	290.59	237.70	211.30
India	-	91.67	66.00	52.80
Colombia	-	83.21	79.30	74.90
Australia	66.04	56.80	26.40	26.40
Other	-	247.27	-	-
World Total	22,946.87	19,534.993	17,335.20	13,101.7

In addition to bioethanol, this microorganism also converts such sugars from agricultural wastes into beer and wine. Bioethanol is used as a car fuel in many countries and therefore the bioethanol production is increasing rapidly (Wyman, 2003). Moreover, bioethanol could be produced from agricultural wastes because wastes do not compete with food production and do not disturb human's food chain. Synthetically, 7% ethanol can be made from petroleum resources and 93% ethanol through fermentation process using microorganisms to convert biomass materials into ethanol (The News, 2009). This production of bioethanol by synthetic and fermentation method was estimated in 1995. Bioethanol is already the largest product of the fermentation industry, with 65 billion litres production worldwide annually. Most of the countries all over the world have established or are establishing programmes to add ethanol to gasoline. To strengthen the economy of rural areas, improve the quality of air and for reduce the demand for oil, many countries look for bioethanol production. For example, the Brazilian government has developed industries and established programmes that support these industries to produce successfully bioethanol from sugarcane. Bioethanol production has reached approximately to 51,000 mill litter across the world. In this production, USA and Brazil are the first producers. In addition, China, India, Malaysia, Singapore and European countries have successfully produced bio ethanol from agricultural wastes (Lichts, 2007, 2008, 2009, 2010). From ethanol produced across the world, 73% is used as a fuel, 17% as beverage ethanol and 10% as industrial ethanol, in average (Table 1).

BIOETHANOL IN PAKISTAN

In Pakistan, bioethanol production from agricultural wastes such as rotten potato, mango peels, beans and banana is under investigation in laboratory scale. Agricultural wastes in Pakistan are not commonly used for biofuel production. This paper therefore aimed to cover several perspectives on the size of the bioethanol feedstock resource in Pakistan. For production of bioethanol, agricultural wastes are also included in feedstock and there are approximately 17.86Â MT of wasted crops in Pakistan that can potentially produce 4.91Â GL of bioethanol per year. The most favourable bioethanol production sources in Pakistan are potato, mango peels, beans and banana. Bioethanol as fuel can replace gasoline with good performance results of engine, and does not pollute the natural environment. Hence, bioethanol fuel is considered environmentally friendly fuel (The News, 2009).

SOURCES OF BIOETHANOL

Waste sugar feedstock

During fermentation, microorganisms convert the fermentable sugars into bioethanol and some other co-products, as well as food. The 6-carbon sugars are typically used by these microorganisms, glucose being one of the most common sugars. Glucose is converted very easily into bioethanol. Sugar containing materials (known as saccharides) include sweet sorghum, sugar beet, banana, mango, water melon, sugarcane (Figure 1) and other fruits; these are examples of sugar feedstock (Kobayashi et al., 1998). The use of these materials for bioethanol production is highly expensive and humans use them as food. Therefore, the wastes of these sugar containing sources can be fermented by using different microorganisms of interest. To ferment these glucose containing wastes materials into ethanol, specific type of

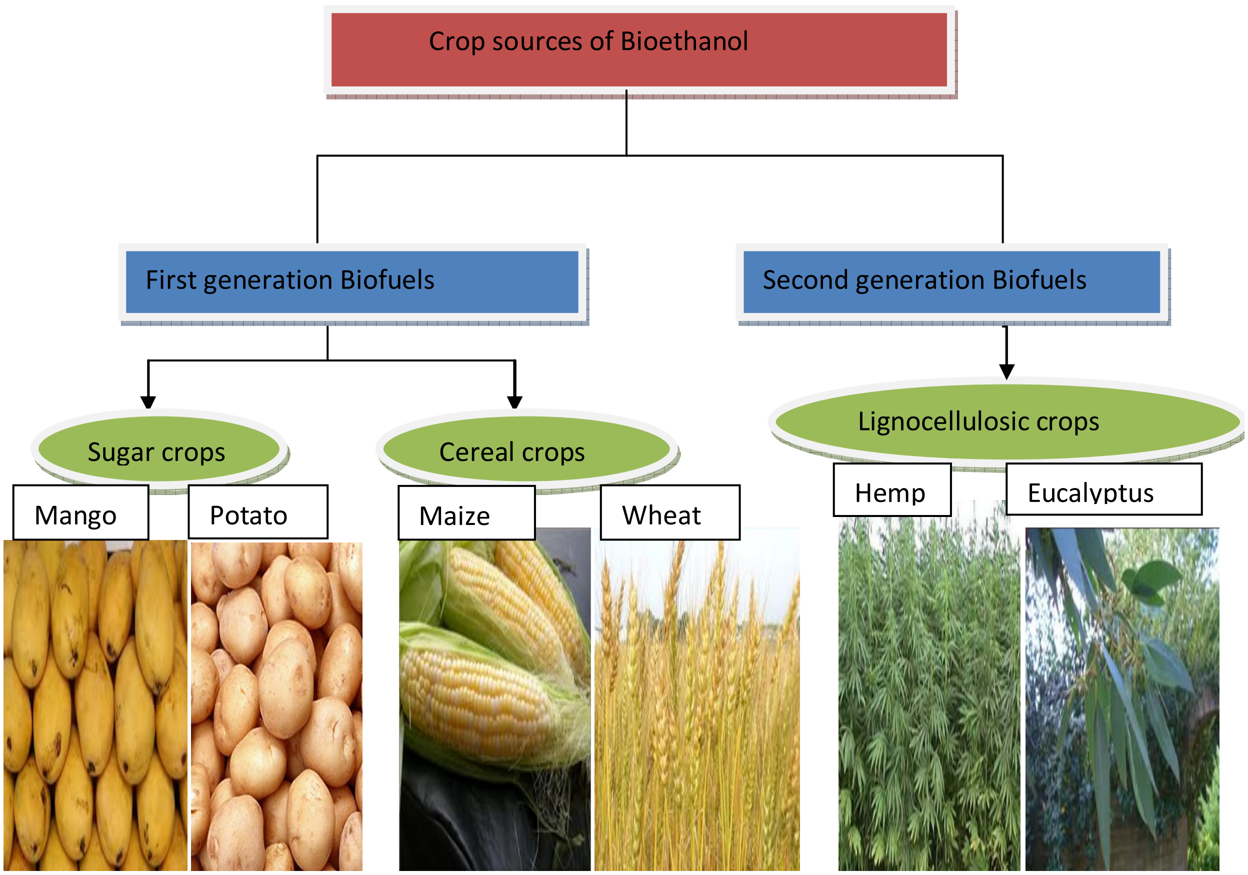

Figure 1. main sources of bioethanol.

yeast, *S. cerevisiae*, which is also known as baker's yeast, is used especially in baking industry. One hundred grams of glucose will produce 270 ml of ethanol theoretically. However, the actual yield of bioethanol produced is less than 100%; the microorganisms use part of the glucose for their growth (Cazetta et al., 2007).

Waste starchy feedstock

Starchy materials are also potential resources for the bioethanol production. Starch molecules are polysaccharides made up of long chains of glucose units covalently linked. Before the fermentation process, the starchy materials are broken into simple glucose molecules after which the simple sugar units are easily fermented by the microbes. Examples of starchy materials used in Pakistan for bioethanol production include cereal grains, potato, sweet potato, beans, cassava and cereal grains. Pakistan is an agricultural country and produces millions of tonnes of wheat and maize. Depending on the process, 30 kg of maize grain can produce 10 to 13 L of pure bioethanol. As these materials are also too expensive and it is included in the human food chain, therefore its wastes are collected from places where it is crushed into flour or from the industries where it is used for various products. So we can get wealth out of wastes. Through liquefaction and saccharification process, these starchy waste materials are hydrolysed by mixing with water and enzymes, and then cooking at 95 to 110°C of temperature to break down the starch into fermentable sugars. The cell walls are ruptured when the slurry of material and water is heated at high temperature and agitated. Specific enzymes that are necessary for the chemical bonds breaking are added during the time of heating and cooling (Rani et al., 2009).

Cellulosic feedstock materials

Cellulosic materials are used for bioethanol production. Examples of cellulosic materials are bagasse, straw, paper, cardboard, wood and materials of plant cellulosic fibrous such hemp, giant reed, eucalyptus tree and miscanthus. Cellulosic resources are immensely widespread and found abundantly everywhere. These cellulosic materials have the potential to be used for the production of bioethanol since they are not commonly used by humans and exist in large amount. Moreover, these materials are inexpensive as compared to the sugar and starchy feed stocks and preferably used for bioethanol production. Cellulosic materials are called lignocelluloses because it is composed of lignin, cellulose and hemicelluloses. The function of lignin in plant is to provide structural strength and support the plants body. Relatively, grasses have fewer amounts of lignin molecules than trees. Unfortunately, cellulose and hemicelluloses molecules are enclosed by lignin molecules which have no sugars units, and thus it is very difficult to reach them. As starch molecules, cellulosic molecules are also polysaccharides and composed of long chains of glucose molecules, but have a different configuration of structure. Therefore, it is very difficult to hydrolyze cellulosic materials because of its structural properties and its encapsulation by lignin molecules.

Similarly, hemicellulose is also composed of long chains of sugar molecules linked together; but in addition to glucose which is a 6-carbon or hexose sugar, hemicellulosic materials also comprise pentoses, 5-carbon sugars. The composition of sugar molecules in hemicelluloses varies widely from one plant to another. To get efficient economic processes, it is very important to ferment 5-carbon sugars into bioethanol as they provide vital percentage of the available sugars. For this purpose, special types of microorganisms have been genetically engineered (Badger, 2002). The genetically modified microorganism that has the ability to ferment both 5- and 6-carbon sugars has been developed by the University of Florida. Generally, bacteria are highly efficient for fermentation relative to yeast, which takes hours. Researchers are trying to develop microorganisms with the ability to efficiently ferment at least part of the sugars molecules present into bioethanol.

ENZYMATIC DEGRADATION

Natural enzymes are proteins that are found in plants and specific microorganisms, which catalyze certain chemical reactions. Agricultural wastes materials of sugar and starchy feedstock are hydrolysed by the addition of enzymes to the slurry. The pH is adjusted to about 5.8 and an alpha-amylase enzyme is added. The slurry is heated in pressurized cooker to 95 to 110°F for 30 to 40 min to reduce viscosity of the materials and to give the alpha-amylase enzyme time to break down the starch into short chain dextrin. After cooling by atmospheric air, a second enzyme, glucoamylase, is added after the cooled mixture is pumped into the fermentation tank and then properly agitated. The glucoamylase enzyme breaks down dextrin into fermentable sugar. These sugar molecules are easily fermented by yeast into ethanol. It takes two to three days to ferment the materials into bioethanol or may vary depending on the source of the materials (Jarl, 1969).

In the case of lignocellulosic biomass, to expose the cellulose and hemicelluloses molecules, some pre-treatment steps are required to break the crystalline structure or remove the lignin molecules. After uncovering cellulose, cellulose enzyme breaks the long polysaccharide chain into sugar units that are then fermented into bioethanol. Depending on the biomass material, either physical or chemical pre-treatment methods may be used. Physical pre-treatment methods may use high temperature and pressure, milling, radiation, or freezing—all of which consume large amount of energy. The chemical pre-treatment methods use a dilute acid pre-treatment and alkaline pre-treatment to break apart and dissolve the crystalline structure of lingo-cellulosic materials. After these pre-treatment, the slurry is detoxified to remove materials and then this slurry is sent to a fermentation tank with enzyme and yeast. In simultaneous saccharification and co-fermentation, both the enzymes and the fermentative yeast are added at the same time, where sugar conversion and fermentation simultaneously occurs. The enzyme requires several days to hydrolyze the materials because of its tough crystalline structure.

Enzymatic processes have some advantages: (1) Relatively, they require low energy, (2) they efficiently carry out the fermentation process and show good results and (3) for the fermentation step, they do not need the construction of expensive containers (Badger, 2002).

CONCLUSION

From our discussion so far, it is clear that Pakistan is an agriculture country with enough agriculture wastes. Therefore, we recommend researchers, economists and the government to use their potency in the production of bioethanol as renewable energy resource in Pakistan.

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