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Utilization of waste ripe Banana, and peels for Bio ethanol production using *Saccharomyces cerevisiae*

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Abstract

Dried and ground peel biomass, ripe waste banana and Hydrolyzed peels of green and red banana used for bio ethanol production by *Saccharomyces cerevisiae* with shake flask culture. The substrate was given with different concentration as 1%, 2.5%, 5%, 7.5% and 10% (w/v) along with 1% inoculum. *Saccharomyces cerevisiae* exhibited maximum yield of ethanol in ripened red banana and their hydrolyzed peels about 1.3% and 0.27% (v/v) in 10% substrate concentration. The minimum yield about 0.02% of alcohol was produced in green un hydrolyzed banana peels with 1% substrate concentration. Conversion of reducing sugar range was 21% (g %) in un hydrolyzed green banana peels and maximum of 76% in red ripened banana as a substrate.

Key words: Bio ethanol, Banana, Waste, Peels, hydrolyzed peels, Green banana

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Introduction

Banana is one of major constitute the principal food resources in the world. These cultures occupy the Fourth world rank of the most significant foodstuffs after rice, corn and milk (FAO 1999; INIBAP 2002). Banana trees are produced in large quantities in tropical and subtropical areas. World production of Musa in 2003 was estimated at 102 million tons, of which about 68% was classified as bananas and 32% as plantains (FAO 2003). Banana is an important fruit crop of the tropical and the sub-tropical regions of the world, mainly grown on the Asian, South American, and African continents. Most of the fruit peels/residues are dried, ground, pelletized, and sold to the feed manufacturers at a low price which is not considered a highly viable proposition (Mamma et al. 2008). As per the FAO statistics, India is the largest producer of banana in the world and accounts for nearly 30% of the total world production of banana. Other major producers include China, Columbia, Costa Rica, Mexico, and the Philippines. Though banana peel is a fruit residue, it accounts for 30–40% of the total fruit weight (Emaga et al. 2008) and contains carbohydrates, proteins, and fiber in significant amounts. Since banana peels contain lignin in low quantities (Hammond et al. 1996), it could serve as a good substrate for production of value-added products like ethanol. In India, ethanol is mainly produced from molasses but its alternative uses, low availability, and a

continuous increase in its demand has led to a search for other suitable feed stocks like fruit residues. Waste is an inheritable consequence of the food industry. As concerns over the environment have increased, the protection of the environment can only become possible where there is sufficient knowledge of the range of activities that can defile the aesthetic of environment. Environmental goods (natural resources and biophysical conditions) have gradually turned into an economic variable its consideration in the context of industry performance leads to redesign the processes of production in line with the so-called environmental technology. Fruits are highly perishable products, currently most of the perishable fruits are lost during their journey through the agric–food chain, due to spillage, physiological decay, water loss mechanical damage during harvesting, packaging and etc., so recent years effort have been directed towards the utilization of cheap and renewable agricultural sources such as banana peels waste as an alternative substrate for production of alternative bio-fuel like ethanol. Hence, the present investigation envisaged utilization of ripened waste banana and banana peels used as a substrate for production of ethanol using *Saccharomyces cerevisiae* has been presented.

Materials and Methods

Preparation of substrate

Ripe waste banana and peels wastes of two varieties such as *Musa ornate* (Royal red) and *Musa robusta* (Lacatan) used as raw substrate were collected from local market and cleaned and chopped disinfected with 70% ethanol. Then this was kept refrigerated.

Organism

Saccharomyces cerevisiae strain was obtained in the form of pellets from Dharanai Sugars and Chemicals Ltd, Vasudevanallur, India. The strain was revived in 100ml of YPD medium with shake flask culture and it was maintained as slant culture. From the slant, culture was developed as inoculum with same YPD medium with 1% of banana. Further 1% of culture (10^6 cells/ml) used as inoculums for production.

Preparation of fermentation medium

Disinfected and crushed banana, peels and hydrolyzed peels were made as different concentration up to 1%, 2.5%, 5%, 7.5% and 10% w/v. with sterile distilled water along with ammonium phosphate in traces as nitrogen source, and then the content was used for fermentation. The medium was inoculated with 1% of 24hrs revived yeast culture.

Estimation of Ethanol (Caputi et al. 1968)

The ethanol content in the fermentation medium was estimated after appropriate incubation time using chromic acid method adopted by Caputi et al. (1968). 10 ml of fermented medium was taken and underwent distillation process. Approximately 5ml of distillate was collected and combined with 25ml of chromic acid and content was made up to 51ml with sterile distilled water and kept in water bath at 80ºC for 15 minutes, after cooling read at 600nm. Thus the obtained values plotted with standard values.

Determination of reducing sugars (Miller 1959)

Reducing sugars in untreated and pretreated raw materials in the culture broth were determined by dinitrosalicylic acid (DNS) method with glucose as standard.

Saccharification of banana peel (Tiwari et al. 1986)

Peels were hydrolyzed at 10 psi for 30, with sulphuric acid at concentration range 1% of Crushed peels, the acid hydrolysates were cooled to room temperature. The solution was neutralized with sodium hydroxide (5% w/v).

Results and Discussion

Red and green banana peels, ripened waste banana, and hydrolyzed peels used for the production of bio ethanol. The substrate concentration used was 1% - 10%. Ethanol was produced in all substrate concentration. Among them, ripened red banana produced higher yield about 1.35% of ethanol in 10% of substrate concentration whereas, ripened green banana yielded 0.99% in 10% substrate concentration (fig-1). It seemed that utilization of reducing sugar also more in ripened banana. Ethanol production on banana peels and hydrolyzed peels concerned was depicted in (fig-2).

Fig 1. Ethanol production using Red Banana, banana peels & hydrolyzed peels

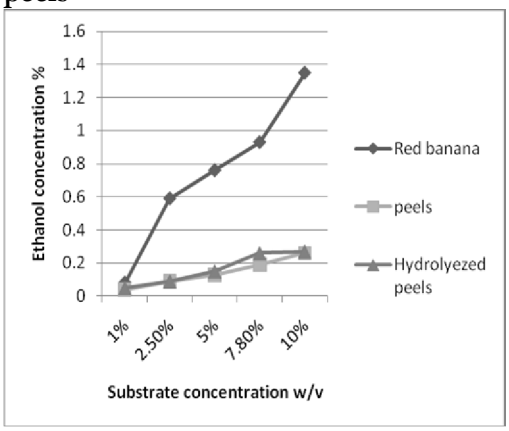
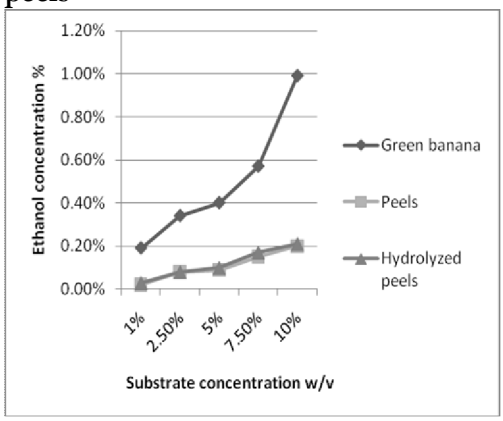


Fig 2. Ethanol production from Green Banana, Banana peels and Hydrolyzed peels



Hydrolyzed red banana peels and un hydrolyzed peels produced the maximum of 0.27% and 0.26% respectively in higher substrate concentration whereas, green hydrolyzed peels and un hydrolyzed peels yielded 0.20% and 0.22% of ethanol in 10% of substrate concentration. These results showed that red banana, peels and hydrolyzed peels produced higher yield than green banana peels and hydrolyzed peels. Regarding the utilization of reducing was also analyzed in this investigation, it revealed that, all substrate concentration s the reducing sugar utilized by the organism. Comparatively the reducing sugar utilization was more in ripened banana than hydrolyzed peels and un hydrolyzed peels. The maximum utilization of reducing sugar was in ripened red banana i.e. about 76.1% and the minimum utilization was happened in un hydrolyzed green banana peel in lower concentration. The overall results stated that the substrate moderately utilized by the organism and the productivity also varied in the medium due to variation of substrate concentration and availability of reducing sugar.

Utilization of substrate during fermentation might have been influenced by various factors such as pectin, complex sugar, hemicelluloses, fiber and lignin. Production of ethanol was very low in un hydrolyzed peels than hydrolyzed peels. Previous reports stated that, the difference in performance of the yeast strains may be due to preferential utilization of pentose and hexose sugar prevailing in the hydrolysatae. The residual reducing sugar also decreased significantly due to the efficiency of the strains in utilization of reducing sugars. The decreased ethanol yield by the strains may be due to increased production of aldehydes, phenols, lactic acid and an increased sensitivity of strains to ethanol tolerance (Olsson et al. 1996). The temperature may be another major factor to influence with the production of ethanol; another report revealed that the temperature and substrate concentration will have influence in yield of ethanol during fermentation process when using different strains of naturally isolated yeast (Manikandan et al. 2007). When banana fruit was hydrolyzed, them as s amount s of H_2SO_4 and NaOH consumed were higher than the ethanol mass produced. These results lead to an economically unviable process, requiring more research to diminish the amount of raw materials used in the process. The high amount of work required by enzymatic hydrolysis when lingo cellulosic material (flower stalk and banana skin)was processed came from the mechanical work consumed in material pretreatment (crushing, shattering and delignification) and the work consumed in stirrers due to the long reaction time ,that is necessary for destroying cellulose chains (Velásquez-Arredondo et al., 2010). This means that research has to be continued aiming to reduce the reaction time in enzymatic hydrolysis.

Fig 3. Utilization of Reducing Sugar in Red Banana, Peels and hydrolyzed peels

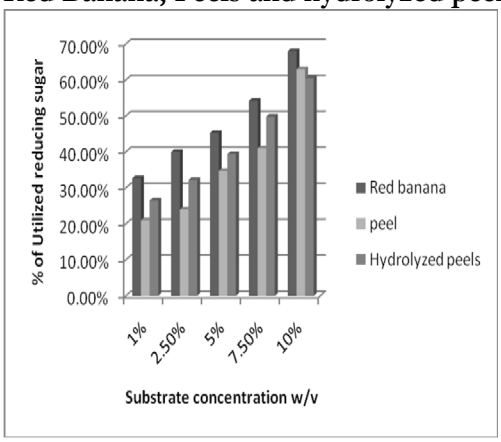
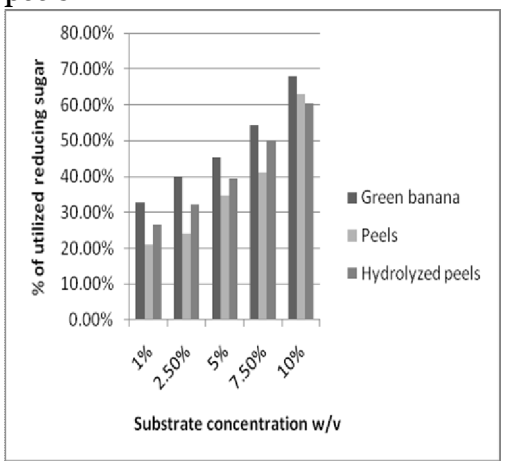


Fig . 4 Utilization of Reducing Sugar in Green Banana, Peels and hydrolyzed peels



The result of this study indicated that industrial yeast with good fermentation attributes, which may enhance ethanol yield and minimize cost of production, could be obtained from ripe banana waste and hydrolyzed red banana peels. Banana peels are always available in abundance in India and thus serve as readily available raw substrate for the production of ethanol. At present ethanol is produced from molasses, which is byproduct of sugar industries. The cost of production increases as the demand for molasses has increased (Ramanathan 2000). So, alternative source like banana wastes, other agriculture wastes could be used for bio ethanol production.

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