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Detection of D-Xylose Activity of Yeast Species Isolated from Local Yoghurt and Sugar Cane Juice in Karachi

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ABSTRACT

Biofuels are vigor transporter that can hoard energy strained from biomass. There are various forms of biomass which are derived from food, wood and fiber; that can be obtain by processing slug of industrial, agricultural, forestry material. About twenty samples of yoghurt, and sugar cane juice were purchased indiscriminately from various places of Karachi. About 10 strains of yeasts were isolated by using Yeast extract peptone agar. Each strain was examining on the basis of cultural, morphological and biochemical characteristics. The total numbers of yeast strains were 10 from yoghurt while 5 from sugarcane extract. These isolates have ability for the production of diverse extra cellular enzymes by fermenting different sources of carbon; in resultant alcohol is produced.

Keywords: *Biofuels, biomass, slug.*

INTRODUCTION

Sugarcane (Saccharum officinarum) is economically is amongst the world's economically chief crops all around the world. Pakistan is at 4th position for the production of Sugar cane. Worldwide it is cultured on 20. 42 million ha with an estimated total manufacture of 1333 million metric tons (Agribusiness Handbook, 2009). Biomass goes through a complex four-part process to generate ethanol, which is used as a fuel in place of or in addition to conventional petroleum products (Preez, 1994). Raw materials, both grain and cellulosic biomass, are first pretreated in order to begin breaking down the material and generating more surface area for the second step, hydrolysis (McMillan, 1994). Hydrolysis is accomplished either by the use of enzymes or chemicals. In this step the complex carbohydrate chains in the biomass are broken down to simple sugars. Finally, these sugars are fermented by microorganisms, yeast, fungi, or bacteria, which produce ethanol in a dilute form. In order to concentrate the ethanol, distillation techniques are used. If pure ethanol is required, the product is subjected to further separation techniques (Visser, 1990). Microorganisms that quickly hydrolyze sugar xylose to ethanol with great output; it is necessary to develop such techniques for the production of ethanol that are cost effective and able to produce at large scale. Certain fungi and bacteria are capable to ferment xylose to ethanol has been recognized for many years (Simon, 2005). A variety of wild-type fungal and bacterial genera can carry out direct fermentation of xylose. Fungal genera, frequently show increase production but undergo from low rates, include Fusarium, Monilia, Mucor, Neurospora, Paecilomyces, Polyporus, and Rhizopus. Furthermore bacterial species includes both from mesophilic and thermophilic genera (Dien, 2003). As the need of ethanol is increasing; it is required that to isolate such strains that produce increase amount of ethanol by using such techniques that require low budget. So it can be available in market at cheaper rate (Sures 1999; Suresh 1999). One of the process is yeast cell immobilization that is prudent as compare

to other commercially available systems, it can provide faster rate of fermentation. The cost is low due to in situ removal of cells (Chaudhary, 1996). It also protects cells from the lethal effects of acidic pH, osmotic pressure, inhibitors, temperature etc which play essential role in high ethanol production at cheaper rate (Sree NK, 2000).

METHODOLOGY

Collection of samples: Samples of yoghurt, and sugarcane juices, were collected randomly from local markets of different areas of Karachi in sterile bottles and kept at 4°C.

Isolation of yeast strains: The samples were serially diluted, plated on a selective medium, yeast extract peptone dextrose agar and were incubated at 28°C for 48 hours. The colonies appeared were further purified.

Morphological characterization: The strains were stained by lacto phenol-cotton blue, carbol fuchsin and seen under phase contrast microscope.

Determination of alcohol producing ability: Table 1: Morphological, Biochemical and Physiological characteristics of the yeast strains isolated from yoghurt Assimilation

RESULT & DISCUSSION

20 samples of yoghurt, and sugar cane juice were monitored for bioethanol production from where 15 yeast strains were isolated and purified (Table I and II). Maximum yeast strains were isolated from yoghurt samples, followed by sugarcane juice. Most of the isolated colonies exhibited smooth surfaces with circular margins. The color of the colonies showed a wide variation of creamy white and pinkish. The cells were found to be of various shapes such as round; oval, spherical and ellipsoidal. (Table I and II). The physiological researches of each yeasts strain were carried out by using over 1 test for assimilation of carbon and their catalase activity. The utilization of xylose was tested. The biochemical analysis of the strains isolated from yoghurt and sugar cane samples showed that all the strains could grow in presence of sugars and urea and ferment them (Table I and II respectively). The surface, margin and color of the colonies isolated from the various samples differed from each other. However, smooth, circular and creamy white colonies were found to be more prevalent. All the strains isolated were found to assimilate the xylose and produce detectable amount of xylanase respectively. The strains also exhibited catalase ability of various degrees. The result of this study indicated that most of the indigenous yeasts, isolated from yoghurt and juice samples showed good fermentation attributes, which might enhance ethanol yield that would contribute for the cost effective role in the production of bioalcohol and enzymes of industrial importance.

Yeast strain	Surface	Margin	Color	Glucose Sugar / Alcohol	Fructose Sugar / Alcohol	Sucrose Sugar / Alcohol	Maltose Sugar / Alcohol	Urea Sugar / Alcohol	D-xylose	Catalase activity
D1	Smooth	Irregular	Creamy White	++/+	+++/+	++/ +	++/ +	-/+	+	+
D2	Rough	Circular	Creamy White	++/+	+++/+	+++/+	+++/+	-/+	++	+
D3	Smooth	Circular	Creamy White	+++/+	+++/+	+++/+	+++/+	-/+	++	+
D4	Rough	Irregular	Creamy White	++/+	+++/+	++/+	++/+	-/+	++	++

Yeast strain	Surface	Margin	Color	Glucose Sugar / Alcohol	Fructose Sugar / Alcohol	Sucrose Sugar / Alcohol	Maltose Sugar / Alcohol	Urea Sugar / Alcohol	D-xylose	Catalase activity
D5	Rough	Irregular	Creamy White	++/+	+++/+	+/+	+/+	-/+	++	+
D6	Rough	Circular	Whitish black	+/+	+++/+	++/ +	++/+	-/+	++	+
D7	Rough	Irregular	Whitish black	++/+	+++/+	+/+	++/+	-/+	++	++
D8	Rough	Irregular	Creamy White	++/+	+/+	+++/+	+++/+	-/+	++	+
D9	Rough	Irregular	Creamy White	++/+	++/+	+++/+	+++/+	-/+	+	+
D10	Rough	Irregular	Blackish	++/+	+++/+	+++/+	+/+	-/+	+	++

Table 1: Morphological, Biochemical and Physiological characteristics of the yeast strains isolated from yoghurt Assimilation

Figure 1

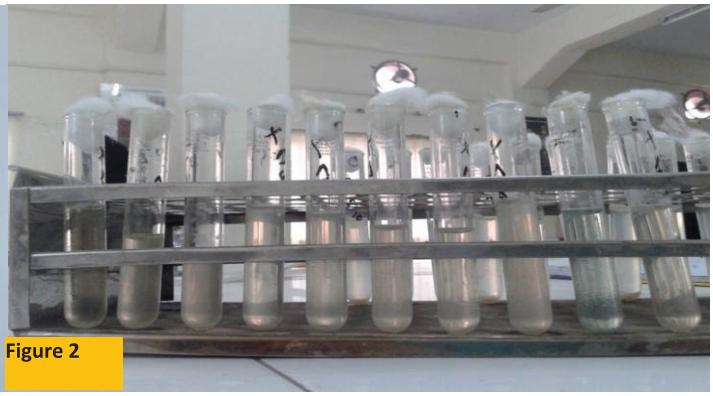

Figure 2

Figure 1: Yeast isolated from yoghurt and sugar cane samples.

Figure 2: Production of alcohol from D- xylose.

Yeast Strain	Surface	Margin	Color	Gluc. Sugar / Alcohol	Fruc. Sugar / Alcohol	Suc. Sugar / Alcohol	Malt. Sugar / Alcohol	Urea Sugar / Alcohol	D- xylose	Catalaes activity
A1	Rough	Irregular	Creamy white	+/+	++/+	+++/+	+/+	++/+	++	+
A2	Rough	Irregular	Creamy white	+++/+	++/+	+++/+	++/+	++/+	++	+++
A3	Smooth	Circular	Creamy white	+++/+	+++/+	+++/+	++/+	+++/+	+++	+
A4	Rough	Circular	Creamy white	+++/+	+++/+	+++/+	++/+	++/+	++	++
A5	Smooth	Circular	Creamy white	+++/+	++/+	+++/+	++/+	+++/+	+	++

Table 2: Morphological, Biochemical and Physiological characteristics of the yeast strains isolated from Sugar cane Assimilation

CONCLUSION

Our aim was to produce bioethanol from yoghurt and sugar cane. There are various techniques available industrially. Though, the leading apprehension of the process is its cost. For this we must use some cost efficient bioethanol production process and in our study we used one of the most cost efficient products for bioethanol production and we hope that some efficient research on these procedures will make them used in daily industrial procedures for bioethanol production.

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