PRODUCTION OF XYLITOL FROM LIGNOCELLULOSE – A REVIEW

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Victor Toluwani Fapohunda[1] and Hafiz Awofe Akinyele[2]
[1,2]Department of Microbiology, Federal University Oye-Ekiti, P.M.B 373 Oye-Ekiti, Ekiti state.
Corresponding Author: hafiz.akinyele@fuoye.edu.ng
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Abstract

Lignocellulose is made up of cellulose, hemicelluloses and lignin. Lignocellulosic substrate can be utilized in the synthesis of certain chemicals and fuels. Cellulose and hemicellulose can be decomposed into sugars, which are the major substrate for fermentation, and through biocatalytic and chemocatalytic pathway to value added products. In this short review, updated account is analysed on various aspects of lignocellulose compounds which can be biodegraded by fungi, yeast and bacteria to produce xylitol. Xylitol is a high value sugar-alcohol produced when D-xylose (from hemicellulose fraction of lignocellulose) is reduced by enzymes and is employed in food and pharmaceutical industries. Xylitol has large number of advantageous properties, such as low-calorie sweetening power and anticariogenicity which justifies the high industrial demand for xylitol. Xylitol is used as an ideal sweetener for diabetic patients. Industrially, xylitol is manufactured by catalytic reduction of pure xylose, which has some disadvantages. This article reviews the literature on the processes for xylitol production and identifies alternative methods for improved xylitol production to compete with the current chemical process.

Keywords: Fermentation. Lignocellulose; xylitol, Xylose, Xylose Reductase

Introduction

Lignocelluloses can be obtained by nature from wood, grass, agricultural waste, forestry wastes and municipal solid waste (Pérez *et al*., 2002).A wide variety of biological and chemical product can be derived from the renewable resources that make up lignocellulose. Disposal problems in the environment arise as a result of decomposition of agricultural waste in large quantity in the environment which leads to environmental deterioration and loss of potentially valuable materials that can be used in manufacturing industries (Sánchez, 2009). A good source of renewable energy are lignocellulose materials which contains approximately 90% dry weight of plant material in form of cellulose, hemicellulose, lignin and pectin stored in it (Kumar *et al.*, 2009). Fractionation to obtain a variety of useful chemicals from the raw materials have been reported by (Moldes *et al*., 2007) including the xylitol production by *Debaryomyceshansenii*. Agricultural products containlignocellulosesas the main organic materials. D-xylose is the major carbon source of forestry products and renewable agricultural residues. Plant tissue on dry basis contains 95% of D-xylose and L-arabinose of arabinoxylan D-xylose and L-arabinose constitute 95% of arabinoxylan hemicelluloses in plant tissues on dry basis and pentosans constitute around 19-33%, 10-12% and about 40% (dry weight basis) in hardwoods, softwoods and agricultural waste, respectively (Winkelhausen and Kuzmanova, 1998). Some sources of xylose from lignocellulose residue include rice straw (Roberto *et al*., 1996), barley straw (Du Preez, 1994), and seed coats of wheat, corn, rice, soybeans, and oats (Whistler, 1993).

Certain fruits and vegetables contain xylitol which is a five-carbon sugar alcohol. Xylitol is most importantly used as a sweetener for diabetic patients (Ylikahri, 1979). Other important uses of xylitol include thin coatings on vitamin tablet, in chewing gum, soft drink as an anti-cariogenic agent in toothpaste formulations, beverages,(Hyvönen and Koivistoinen, 1982),(Mäkinen, 1992). The pharmacological properties of Xylitol, its high sweetening power, its anticariogenicity and its insulin-independent metabolism has made xylitol to receive global demand. More than 50 countries have Approved the use of Xylitol in foods, pharmaceuticals, and oral health products (Povelainen, 2008). Chemical hydrogenation of pure xylose can be used to produce Xylitol or by biotechnological method.

Many extensive researches have been carried on xylitol because of its growing market and high added value (Parajo, 1998). Chemical reduction of pure xylose in the presence of a nickel catalyst at high temperature and pressure is the method currently used in the production of xylitol in the industry (Kitpreechavanich *et al*., 1984). About 50–60% of xylitol is produced from the xylan fraction and due to complex purification procedure the resultant product is expensive (Parajo*et al*., 1998). Due to the difficult process, high cost and energy intensive of the chemical process, alternative methods to conventional processes are proposed. The fermentation process and the enzymatic process are two biotechnological processes that seem promising. High-quality and cost-effective product through biotechnological process are highly attractive alternatives. Bacteria, fungi, and yeast are been used for xylitol production from xylose or hemicellulosic hydrolysate in the fermentation process. Among the microorganisms investigated yeasts are considered as the best xylitol producers. In the fermentation process, production of xylitol obtainable from xylose is in a range of 65–85% of the theoretical value. Due to some preparatory activities, such as sterilization and regular inoculum development, the application of the fermentation process on industrial scale is time consuming. The major advantage of the fermentation process over chemical procedures is its lower cost due to the non-necessity of xylose purification (Parajo *et al*., 1998). However, the fermentation process has not yet been able to accumulate the advantages of the chemical process. Production of xylitol from xylose through the process of enzyme technology can be an attractive alternative to both fermentation and chemical processes. Compared to the fermentation process, the enzymatic approach employs xylose reductase (XR) for xylitol synthesis and is expected to obtain a substantial increase in productivity compared to the fermentation method. Reports on the enzymatic conversion of synthetic xylose to xylitol using XR are scarce (Melaja and Hämäläinen,

1977). There is limitation in the application of xylitol as sweetener due to its high price despite a variety of its uses. To lower the production costs a lot of researchers are working towards the development of improved technologies. Enzymatic approach to xylitol production from xylose present in the lignocellulosic biomass may provide an alternative for the chemical process in this field (Neuhauser *et al*., 1998). This review describes the current literature on the processes involved in the production of xylitol, both the chemical and biotechnological processes, the microorganisms involved and to identify ways to improve enzymatic xylitol production.

Production of Xylitol from Hemicellulose Hydrolysate

Cellulose, hemicellulose, and lignin are the main parts of lignocellulose. The complex structure of lignocellulose in plants prevents it from cell destruction by bacteria and fungi. Cellulose and hemicellulose must be hydrolyzed into their corresponding monomers (sugars) for easy utilization by microorganisms through the process of fermentation. The main component is xylan of hemicellulose fraction of hardwoods and agricultural residues. Xylan is a polymer made from xylose units that can be hydrolyzed to this sugar by mineral acids or xylanase. Under specific conditions, the solid residue from hydrolysis of acid contains both the cellulosic and lignin fractions that can be separated into other fractions and utilized for different product applications. The hydrolysis of biopolymers can be performed enzymatically but most fermentation studies have focused on hydrolysates derived from hydrolysis. Due to hemicellulose relatively low degree of polymerization and heterogeneous structure it is easier to hydrolyse compared to crystalline cellulosic components of biomass (Magee and Kosaric, 1985). Acid can hydrolyse the lignin (phenolic fraction), that remains as a solid residue in acid medium both of cellulose and hemicelluloes. Hemicelluloses are more susceptible than cellulose to the hydrolytic action of catalysts due to their open and branched structure. Carbon source for microorganisms is hydrolyzed from polysaccharides of raw material (lignocellulosic substances) to the corresponding sugars through fermentation processes. The most commonly employed catalysts for fermentation of hemicellulose are H_2S0_4 and HCl are the most commonly employed catalysts for fermentation. The liquid phase (containing xylose, byproducts, and compounds derived from other fractions of the raw materials such as extractives or lignin) prepared by hydrolysis can be used for making fermentation media suitable for xylitol production. Xylitol production is influenced by important factors like defined media formulated by chemicals according to biocatalyst requirement, concentration of xylose as substrate such that increased concentration of xylose results in improved yield and productivity. However, when lignocellulosic hydrolysates are involved in making culture media, some additional effects related to the concentration of substrate must be considered. By increasing concentration of xylose using evaporation an inhibition of microbial metabolism and reduction of cell growth occurs which is dependent on a simultaneous increase in the concentration of other non-volatile compounds (Olsson and Hahn-Hagerdal, 1996). Minerals or metals resulting from the corrosion of the equipment or ions contained in the lignocellulosics, compounds like furfural and hydroxyl methyl furfural derived from degradation of sugars at high temperatures or acetic acid liberated from the acetyl groups in biopolymers, chemicals like aldehydes, phenolic compounds and aromatics which are derived from lignin degradation and compounds derived from extractives are different groups of inhibitors in hydrolysates from acid catalysis. Some organic acid like syringic, caproic, caprylicvanillic, pelargonic and palmitic acids are other important and effective inhibitory compounds found in hydrolysates. The main parameters that determine the maximum allowable concentration for each process are some factors such as the microorganism utilized, adaptation potential of the microorganism, mechanism of fermentation process employed, and the simultaneous presence of several other inhibitors.(Parajo *et al*., 1998), (Mussatto *et al*., 2005). Acid hydrolysis should be carried out in a way that allows: high concentration of xylose as much as possible, concentrations of inhibitory byproducts in the ranges should be tolerable by the microorganism, and high selectivity towards cellulose degradation in order to make hydrolysates appropriate as fermentation media (Parajo *et al.,* 1998).

Processes for producing xylitol

Chemical process

The reduction of pure xylose obtained from hardwood or hemicellulosic hydrolysate in the presence of a Raney nickel catalyst is the process through which xylitol is manufactured in the industries. The extraction of xylose from hemicellulose by acid-catalyzed hydrolysis starts the chemical production of xylitol. Xylose-rich hemicellulosic hydrolysate can be utilized for xylitol production through hydrogenation of xylose at 80–140°C and hydrogen pressures up to 50 atmospheres in the presence of metal catalysts (Raney nickel) after the color is removed and the xylose is purified. Chromatography is used further to purify the xylitol solution formed, and then concentration and crystallization of the product to obtain pure xylitol (Winkelhausen and Kuzmanova, 1998).The xylitol production process is costly due to the extensive separation and purification stages and the yield of xylitol is only about 50–60% of the xylan fraction (Parajo *et al*., 1998).

Biotechnological processes

Fermentation process

The production of xylitol from commercial pure xylose or hemicellulosic hydrolysate by fermentation process utilizes bacteria, fungi, and yeast. The production of xylitol using bacteria and fungi has been studied to a lesser extent compared to that of yeast. Xylitol is produced by a few bacteria such as *Enterobacterliquefaciens* (Yoshitake *et al*., 1973) *Corynebacteriurn sp*. (Rangaswamy and Agblevor, 2002), and *Gluconobacteroxydans* (Suzuki *et al*., 2002). Studies of xylitol production from D-xylose using filamentous fungi are very few (Gong *et al.,* 1981). Among the microorganisms yeasts are considered to be the best xylitol producers. As a result, yeasts have been studied extensively in the last few decades by several researchers (Gong *et al*., 1981), using high cell densities and a defined medium under aerobic conditions. *Candida guilliermondii* and *C. tropicalis* were found to be the best xylitol producers, the yeasts produced 77.2 g l^{-1} xylitol from 104 g l^{-1} xylose. *C. tropicalis* produced xylitol at a yield of 77–80% of theoretical value (0.91 g g-1) in a medium containing 100 g l^{-1} D-xylose. The best xylitol producers belong to the genus *Candida* which was confirmed by the screening of different xylose-

assimilating yeast. In the fermentation process using yeast, the yield of xylitol obtainable from D-xylose is in a range of 65–85% of the theoretical value. Certain factors limits the production of xylitol through the fermentation process, such as precise control of culture conditions, expensive nutrients, huge water consumption, and the type of process (Sampaio *et al*., 2008). Thus, the application of the fermentation process on an industrial level is time-consuming, being associated with some preparatory activities such as sterilization and regular inoculum development involving input of energy, labor, and time, leading to decreased productivity. The advantage of the fermentation process over chemical procedures is its lower cost due to the non-necessity of extensive xylose purification is the advantage of the fermentation process over chemical procedures (Parajo *et al*., 1998). The viability of fermentative xylitol production has been studied as an alternative dependent on the optimization of the various fermentation variables such as nutritional composition (substrate, nitrogen source, and micronutrients), the culture and process conditions, as well as the genetic nature of the microorganisms (Prakasham *et al*., 2009).

Enzymatic process

An alternative and promising approach forthe production of xylitol from xylose is by using enzyme technology. The enzymatic conversion of D-xylose into xylitol using xylose reductase (XR) of *Candida pelliculosa* coupled with the oxidoreductase system of *Methanobacterium sp.* has been reported by (Kitpreechavanich *et al*., 1984). Stoichiometrical conversion of xylose to xylitol with an equivalent consumption of NADPH was observed by the authors

Solid-liquid extraction

Xylitol is found naturally in fruits and vegetables, as well as in yeast, lichens, seaweed, and mushrooms. Solid-liquid extraction can be used to extract these sources, but its small quantity in the raw materials (less than 900 mg/100 g) is a major economic drawback for this method (Pepper and Olinger, 1988). Extraction method, chemical method and biotechnological method can be used to produce xylitol

Chemical production of xylitol

Acid-catalyzed hydrolysis of xylan to produce xylose start the chemical production process (Counsell, 1977). However, the presence of other polymers different from xylan in the hemicellulosic residue of the biomass produces hydrolysates produced from the presence of other polymers different from xylan in the hemicellulosic residue of the biomass has various impurities including glucose, arabinose, galactose and mannose because of breakdown of related polymers. Expensive purification steps are required when pure xylose is important (Hyvönen and Koivistoinen, 1982). Monosaccharides residue that cannot be removable by ion exchange chromatography and activated carbon are considerable part of the impurities (Winkelhausen and Kuzmanova, 1998).Color, metal ions, and other impurities, the xylose-containing hydrolysate is then hydrogenated at 80-140°C and hydrogen pressures of around 5000 kPa in the presence of metal catalysts after the purification procedure and the removal of proteins. The xylitol solution is produced through reduction method, chromatographic fractionation used in purification of the solution and the product is then concentrated and crystallized to obtain pure xylitol (Hyvönen and Koivistoinen, 1982). Purification and separation are the most expensive steps. 50-60% of the initial xylose yield xylitol (Winkelhausen and Kuzmanova, 1998). High pollution levels and waste-treatment concerns are the major drawbacks of conventional production method which has inspired researchers to find alternatives for xylitol production and one of the most interesting procedures is microbial production (Winkelhausen and Kuzmanova, 1998).

Microbial production of xylitol

Microorganisms that are able to utilize xylose to produce xylitol is synthesized from D-xylose by NADPH-dependent xylose reductase as a metabolic intermediate (Saha and Bothast, 1997). Xylitol can be manufactured by a number of yeasts and moldsbecause they possess the enzyme xylose reductase. Some of the yeasts with xylitol production capability are *Candida guilliermondii, Candida tropicalis, Candida pelliculosa, Candida boidinii*, and genera of *Saccharomyces, Debaryomyces, Pichia, Hansenula, Torulopsis, Kloeckera, Trichosporon, Cryptococcus, Rhodotorula, Monilia, Kluyveromyces, Pachysolen, Ambrosiozyma*, and *Torula*(Saha and Bothast, 1997). Xylitol can also be produced bySome bacteria species such as *Enterobacterliqufaciens, Corynebacterium sp*., and *Mycobacterium smegmatis* (Horitsu *et al*., 1992). The conversion of D-xylose to xylitol by microorganisms has been extensively studied in yeasts compared to the other microorganisms and it is important for industrial production. D-xylose is converted into xylitol as an intermediate material (by xylose reductase activity)in the biochemical pathway which is to be transformed to xylulose or directly converted to xylulose. Then, xylulose-5-phosphate is generated by the activity of xylulose kinase and goes through pentose phosphate pathway.

Production of xylitol by bacteria

Xylose isomerase enzyme is utilized by most bacteria to convert xylose into xylulose. It is then phosphorylatedto D-xylulose- 5-phosphate (a common intermediate in the prokaryotes and eukaryotes' metabolism)in the second phase, by xylulokinase. Xylulose-5 phosphate phosphoketolase can convert D-xylulose-5-phosphate into glyceraldehyde-3-phosphate and acetyl-phosphate. Similar to the metabolism of glucose by yeasts, this step produces an intermediate product of glucolysis without production of nicotinamide adenine dinucleotide phosphate (NADPH) (Evans and Ratledge, 1984).Xylose can be reduced into xylitol by xylose-reductase and other oxido-reductive enzymes in bacteria, with further oxidation to xylulose(Yoshitake*et al*., 1973).

Production of xylitol by molds

Low yield of xylitol in fermentation with *Mucor sp*. was observed by (Gong *et al*., 1981) on a hydrolysate from sugarcane bagasse hemicellulose.

Production of xylitol by yeasts

An oxido- reductive transformation consisting of two consecutive reactions are used by some strain of yeast totransform xylose into D-xylulose.In the first stage of the reaction, D- xylose is transformed into the intermediate xylitol with xylose-reductase (XR), in the presence of nicotinamide adenine dinucleotide (NADH) or NADPH (Taylor et al., 1990). In the second stage of the reaction, xylitol is oxidized into D-xylulose by either NAD+- linked or NADP +- linked xylitol dehydrogenase (XDH) (Girio*et al.*, 1994). A redox imbalance occurs when oxygen levels are too low as the NADH produced in the xylitol

dehydrogenase step cannot be re-oxidized back to nicotinimide adenine dinucleotide (NAD). Production of NAD+ is limited due to a decreased respiration rate as a result oflow-oxygen induced imbalance. Therefore, the alternate ethanol route is selected at the end of the pentose phosphate pathway. Since the pentose phosphate pathway is still active, NADP + conversion to NADPH continues (Hahn-Hagerdal*et al*., 1994). The accumulation of xylitol is improved bythe resulting redox imbalance in the XR/XDH system, due to the limited supply of NAD + for utilization by XDH. Semi-anaerobic conditions were essential to lower the NAD/NADH ratio in the oxidation step of xylitol to xylulosefor higher xylitol accumulations in D. hansenii. Accumulation of xylitol is as a result of non-regeneration of NAD + in oxygen-limited conditions and its subsequent excretion into the medium (Saha and Bothast, 1997). Under anaerobic and oxygen-limited conditions yeasts with XR activity linked to both NADH and NADPH, like Pichiastipitis, can regenerate the NAD+ consumed in the second step of the xylose metabolism. Therefore, no xylitol accumulation occurs between the cofactors of XR and XDH and the major product in this situation is ethanol. Xylitol can be produced as the major product by yeasts, like *Debaryomyceshansenii*, that consume xylose by XR activity, which is only dependent on NADPH (with complete absence of NADH-linked XR) in the first step of the xylose conversion (Girio*et al*., 1994). In the presence of NAD+, xylitol is oxidized to xyluloseIn the second step,(Vandeska*et al*.,1995). For selecting xylitol-producing microorganisms the presence of either high XR or low XDH activities is considered as a criterion (Parajo*et al*., 1998).

Parameters of Fermentation

Fermentation stage as the most important stage in xylitol production is influenced by several factors influence Fermentation stage which is the most important stage in xylitol production which includes substrate concentration, carbon source, salts and nitrogenous compounds, inoculum, aeration rate, temperature and pH that are explained in the following sections:

Xylose concentration

For yeast growth and fermentation substrate concentration (D-xylose) has been proven to be a very vital parameter. Xylitol is produced in the presence of D-xylose and for the activities of xylose reductase and xylitol dehydrogenase as well as xylitol formation in yeasts, combination of aeration with xylitol concentration is an important and determinative factor.

Production by microorganisms can be affected bythe initial xylose concentration. Increase in xylitol output and the rate of production is as a result of increase in xylose concentrations in the cultures of microorganisms which are able to tolerate high sugar and higher osmotic pressure. Unless the aeration rate is increased, increase in initial xylose concentration will usually lead to decrease in growth rate(Nolleau*et al*., 1995).(Horitsu*et al*.,1992) investigated the effect of D-xylose concentration on the production of xylitol for C. tropical is by varying the concentration from 100 g l-1 to 300 g l-1. At D-xylose concentration of 172 g l-1 the maximum yield of xylitol was obtained. (Walther *et al*., 2001) reported Canadidatropical is ATCC 96745 cells grew vigorously at high initial xylose concentrations and high aeration rate, at the beginning of fermentation and production rate was improved. However, at lower initial xylose concentration and high levels of dissolved oxygenxylitol yield was reduced. They also found that at extremely high initial xylose concentrations the yield decrease which is attributed to the osmotic stress that could be induced in the microorganism at high concentration of sugar in the medium. Desirable xylitol yields could be as a result ofmanipulation of initial substrate concentration and aeration.

Carbon source

According to studies, in the production of xylitolcarbon sources rather than xylose are important. For C. tropicalis cultivationD-xylose wassupplementedwith D-glucoseby (Yahashi*et al*.,1996). D-glucose was utilized for cell growthat the initial stage of fermentation and then D-xylose was consumed. By the addition of glucose to the fermentation media xylitol yield and productivities increased 1.2 to 1.3 times. This condition yielded 104.5 g l-1 xylitol in 32 h and a yield of 0.82 g g-1 xylose. However, the yield of the product was decreased from 0.66 g g-1 to 0.45 g g-1in an investigation by (Silva*et al.*, 1996) for *Candida guilliermondii* FTI 20037, by the addition of glucose to the fermentation medium. In a research by (Yahashi*et al*., 1996) using a recombinant S. cerevisiae, fed-batch fermentation was proposed to overcome this problem. To achieve effective xylitol production glucose concentrations in the medium should be very low for Candida tropicalis (Walther *et al*., 2001). Aerobic conditions should be adopted to reach higher yields and productivities in fermentation media containing substantial amount of glucose, while microaerobic conditions improves yield of xylitol in the absence of glucose. In contrast to glucose as the co-substrate, arabinose appears to be an inducer of xylose reductase enzyme in contrast to glucose as the co-substrate, in that high arabinose concentrations enhanced both yield and productivity of xylitol in the experiments.

Nitrogen source

An important nutrient used as an organic source of nitrogen is yeast extract for some xylitol- producing yeasts, while for some other yeasts has no significant effect on xylitol formation. For better yield These yeasts prefer urea or urea and Casamino acids (Winkelhausen, and Kuzmanova, 1998).

In microorganismsxylitol production and xylose usage is influenced by the nature and concentration of the nitrogen source, the yeast strain is theeffective factor on the two parameters mentioned previously (Parajo*et al*.,1998). Xylitol yield increase was observed by (Vandeska*et al*.,1995) inwhen the fermentation medium was supplemented with urea compared to those supplemented with ammonium sulfate

Inoculum age and concentration

The age of inoculum influence the rate and output of fermentation which also affects the metabolism and viability of the cells (Parajo *et al.*, 1998). In a study by (Pfeifer *et al.*,1996) *C. guilliermondii* FTI 20037 was used to improve the yield of xylitol by inoculum age, inoculum level (concentration) and hydrolysate composition. In this study, xylose concentration in the hydrolysate, inoculum level and inoculum age varied from 37.6 g l-1 to 74.2 g l-1, 0.1 to 6.0 g l-1 , and 16 to 48 hours , respectively. Maximum xylitol yield of 0.74 g g-1 and productivity of 0.75 g l-1h-1 was reached for 3.0 g l-1 of 24-h old inoculum at an initial xylose content of 54.5 g l-1. Undesirable results was obtained (Pfeifer *et al*., 1996) for xylitol productivity and cell growth from *C. guilliermondii*inocula younger than 15 h or older than 24

h and the best conditions were achieved in the aforementioned range (Pérez *et al*., 2002) examine the influence of initial cell concentration of Candida sp. B-22 on xylitol production from D-xylose and found that the rate of xylitol formation was linearly increased and the fermentation time was dramatically reduced with initial concentrations in the range of 3.8 to 26 g l-1 of inoculum. Aeration rate Due to the diversity of effective parameters and the wide range of aeration levels considered in the literature, a detailed study of the influence of aeration on xylitol production is difficult to perform. The optimum condition for xylitol yield and productivity is provided by the adoption of intermediate values of the aeration rate. For example, under low aeration conditions (4-22 mmol l-1 min-1),D. hansenii shifts its metabolism towards xylitol production leading to the highest productivity at oxygen transfer rate (OTR), higher than 2 mmol l-1 h-1. The shift to anaerobic conditions stopped both xylose consumption and metabolic activity which confirmed that oxygen is an essential component for xylose uptake in the study. In a related study, the maximum xylitol yield for Candida tropical is under semi-aerobic conditions, was 0.62 g g-1 xylose, while under microaerobic conditions, the maximum yield was 0.36 g g-1 substrate. Higher yields and productivities were obtainable under aerobic conditions in a medium containing glucose, while in the absence of glucose microaerobic conditions improved yields. These results can be attributed to increased oxygen demand by the high cell densities achieved in the presence of glucose (Walther et al., 2001). 30°C has been reported to be the most suitable temperature for xylitol production by yeasts. With C. tropical is DSM 7524 small temperature variations above this level, do not significantly affect xylitol yield. The xylitol yield was not dependent on temperature, when the cells were cultured in a temperature range between 30°C and 37°C but at temperatures higher than 37°C the xylitol yield decreased drastically. Similarly, xylitol formation in *C. guilliermondii* FTI 20037 decreased with increase of temperature up to 40°C but was the same at 30 and 35°C, (Barbosa, 1988). The optimal initial pH for fermentation depends on the yeast employed. 5.5 and 4- 6 are the optimum pH for Debaryomyceshansenii and Candida sp. respectively. 4.5-5, 6.0 and 7.0 are the best pH for *C. parapsilosis* , *C.guilliermondii*and *C. boidinii* respectively. The pHdrops during the fermentation process if it is not controlled, therefore, in such conditions, the initial pH values have to be higher than under controlled conditions. For example, the optimal initial pH value for *C. boidinii* under controlled condition is 5.5; while with no control, initial pH should be 7.0 (Winkelhausen and Kuzmanova, 1998).

Conclusion

Production of xylitol is becoming more important due to the high demand for the products because of its nutritional and pharmaceutical importance. It is manufactured in the industry for oral hygiene, pharmaceuticals, cosmetics and food sweeteners (baked goods, jams, gelatins, chewing gum, ice cream, etc.). Xylitol can be used in the prevention or treatment of diseases such as diabetes and obesity.The production of xylitol using XR from xylose from yeast is an attractive alternative to chemical and microbial processes. The usage of XR is an alternative of economic interest to the chemical reduction of pure xylose and the fermentation of xylose present in the hemicellulose hydrolysate. The production of xylitol through the chemical process is costly due to difficult separation and purification steps. On the other hand, the fermentation process on an industrial- scale is not feasible due to low yield of product. It is important to maximize alternative methods for the effective production of xylitol using XR enzyme. The enzymatic method might be able to overcome the disadvantages of the chemical process that is largely being used at present and also the fermentation process that is still undergoing research. Research on the application of cheap substrates, the development of multipurpose microorganisms capable of tolerating extreme working conditions and the regulation of processes may make production xylitol economically feasible. Current and future research trends are targeted towards the developments and usage of engineered organisms to tackle the challenges encountered from using conventional naturally occurring organisms.

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