INDIAN INSTITUTE OF TECHNOLOGY GUWAHATI
SHORT ABSTRACT OF THESIS

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SHORT ABSTRACT

Xylitol is a high-quality polyalcohol, mainly used in pharmaceuticals, hygiene products, and food due to its functional properties such as anticarcinogenic, antibacterial, low-calorie, and hypoglycemic properties. At present, xylitol is primarily produced through chemical hydrogenation of xylose at high temperatures (150 °C) and pressure (5.5 MPa) on the reaction with metal catalysts such as Pl, Ru, Pt and Raney nickel. Separation and purification of xylitol is very expensive through this process. However, compared to this method, xylitol production through bioconversion of hemicellulosic hydrolysate by micro-organisms is an environment friendly, less energy-intensive, renewable, and overall economical process. This process ensures high safety, low production cost and high product selectivity. The primary objective of this research is to utilize the agricultural bio-waste (such as areca nut husk) as a feedstock for the production of xylitol, preferably in a repetitive batch fermentation process, with C. tropicalis as the fermentative microorganism. Primary processes include, biomass characterization and pretreatment (Mainly dilute acid hydrolysis and lime treatment), Acidic and Enzymatic hydrolysis of biomass, detoxification of acidic hydrolysate by using activated charcoal and cation-anion exchange resins, detoxified hydrolysate was fermented by C. tropicalis for xylitol production and finally downstream process was performed for product purification. Lignocellulosic materials are inexpensive and readily available biomass in the form of either agricultural wastes or forest residues. These materials can be used as energy producer sources for solids (xylitol, etc.) liquids (ethanol, butanol, etc.) and gaseous (CO, H2, etc.) as energy to meet increasing energy demands. Biomass pretreatment is a predetermined step to fragment lignocellulosic biomass into its basic components such as lignin and carbohydrate molecules. The first objective of present study is the pretreatment and characterization of lignocellulosic biomass namely Areca nut husk (Arecan catechu), which is widely available in the region of North-Eastern part of India. The study includes several physical characterizations like ultimate and proximate analysis, thermogravimetric analysis, crystallinity and chemical characterization that embraced Raman spectroscopy and FTIR. This study revealed that the Areca husk fiber contained 29.17% hemicellulose. Combination of all these properties revealed that Areca nut husk can be explored as the impending potential for low-cost source of xylose.

Direct biotransformation or saccharification of areca nut husk is very difficult because of recalcitrant nature of lignocellulose. To make biomass more amenable for saccharification, lime and acid pretreatment was conducted. But, before conducting the saccharification, compositional analysis was performed to know the initial available constituents in feedstocks. To improve the enzymatic hydrolysis of areca nut husk, various saccharification parameters like xylanase enzyme loading was varied. Results exposed that a highest yield (g/g) of reducing sugar about 90%, 83% and 15% were achieved for Acid Treated Husk (ATH), Lime Treated Husk (LTH) and Raw Husk (RH), at enzyme loading of 15.0 IU/g. Saccharification was performed at a substrate loading of 2% (w/V), 100 rpm agitation, 30 ◦C hydrolysis temperature for 12 h hydrolysis time at pH 4.5 to 5.0. Raman spectroscopy, Zeta potential, x-Ray diffraction (XRD), Field Emission Scanning Electron Microscopy (FESEM), Thermogravimetric Analysis (TGA), and Fourier Transform Infrared Spectroscopy (FTIR) were performed to know the structural changes in native, pretreated and saccharified residues. Structural analysis showed that major part of partial crystalline and amorphous cellulose in the areca nuthusk biomass were hydrolyzed during saccharification. But, hydrolysis results the removal of amorphous substances, disruption of crystalline structure, and the transformation of crystalline zone to amorphous zone were noted. Dilute acid hydrolysis is one of the most promising way for xylose production, which can be further use for xylitol production. But, detoxification is necessary before the fermentation of hydrolysate, because various toxic components present in the hydrolysate causes inhabitation problem. Various detoxification was performed and their effect on sugar loss and inhibitor removal was estimated. Detoxification includes series of treatments, primarily pH adjustment, activated charcoal treatment and Amberlit ion exchange resin treatment. Detoxification removed about 99% of toxic components present in the hemicellulosic hydrolysate. Further, fermentation parameters were optimized by using RSM and ANN techniques. The results of the ANN model showed that it was significantly more robust and accurate in estimating the values of the dependent variables compared to the RSM model. ANN has also been shown to be very accurate in finding optima and predicting optimal yields. RSM is recommended for modelling new processes, while ANN is better suited for modelling nonlinear systems with non-second-order interactions. Finally, Submerged fermentation process was performed by C. tropicalis for production of xylitol under the optimized conditions, which resulted the optimum yield of 0.74 g/g. It was concluded that detoxification by pH adjustment, activated charcoal and ion exchange resins are considered to be the most economical technique for removing toxic compounds from hemicellulosic hydrolysates and this medium is considered to have as huge potential for xylitol production. Downstream processing of xylitol is one of the most important steps during xylitol production by microbial routes. The present research also highlights the product purification and separation from fermented broth. Xylitol crystals were separated by crystallization from fermented broth, accompanied by characterization of xylitol crystal such as X-ray diffraction (XRD), Differential scanning calorimetry (DSC), Thermogravimetric Analysis (TGA) and Raman analysis. DSC analysis revealed that the melting process of commercial xylitol crystals after crystallization (CXCAC) was observed at 93.71 °C with enthalpies of 210.7 J/g However, almost similar results were observed for xylitol crystals from fermentation broth (XCFB), having a melting point at 91.73 °C with enthalpies 220 J/g.