

Pretreatment of pine needle waste biomass through enzymatic cocktail for bioethanol production

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Abstract

Pine needles are one of the most important forest biomass material available with serious implications on local understory vegetation besides incidences of forest fires which have both short and long-term effects on environment. The biomass can be put to varied uses by proper exploration of its constituent biopolymers such as cellulose, hemicellulose, lignin etc. Prior treatment of this lignocellulose biomass through various physio-chemical agents like acids, bases, high energy radiations etc help in faster degradation of these polysaccharides to fermentable sugar and finally to bioethanol. However, various inhibitors; like 5-hydroxymethylfurfural (HMF) and 2-furaldehyde (furfural) released in the hydrolysate affect growth of the fermentative agents due to cellular damage during fermentation. In view of this, environment friendly and economically feasible technologies are required to be experimented/invented for inhibitor free fermentation processes. Enzymatic pretreatments offer several added advantages over chemical treatments with higher conversion efficiency, the absence of substrate loss and use of process friendly operating conditions. Use of three different enzymes i.e. cellulase (8.56 U/mg protein), xylanase (95.19 U/mg), and ligninase (75.24 U/mg) from *Bacillus* spp. CPB-21 & XPB-11, *Pseudomonas* sp. LPB-06 respectively resulted into 35 ml of fermentable sugar without any side product or inhibitor with initial 50 g of pine needles in 50 ml (0.1 M sodium citrate buffer pH 7.0) after 24 hr incubation under continuous agitation. The process scale-up when tried with 1000 ml of hydrolysate using *Saccharomyces cerevisiae*, resulted in about 54 % conversion after 48 hr of incubation with 90% purity. The outcome of the present work seems promising and can help to provide a new environment friendly enzymatic pretreatment process for the production of bioethanol a renewable source of energy from the forest waste after further R&D and scale-up trials

The over-exploitation of our planet's resources has worsened our environment, which is nowadays suffering from climate change more than ever. Elevated gas emissions, the greenhouse effect, and global warming have all contributed to the search for renewable sources, which are in harmony with world's energy needs. Lignocellulosic biomass is a sustainable alternative that produces new-generation bio-based chemicals, such as biofuels, food additives, enzymes, and others [1–3]. Lignocellulosic biomass includes all kinds of agricultural wastes, forestry residues, and feedstocks, as well as marine algae, and it can be provided on a large-scale platform from all kinds of materials [4,5]. In general, lignocellulosic biomasses consist of lignin,

cellulose, and hemicelluloses, some organic extracts and inorganic components, which are turned into ash after combustion. All those components make lignocellulosic biomass a complex group of polymers that are naturally recalcitrant to enzymatic conversion. Lignocellulosic biomass materials are constituted of renewable substrates used for bioethanol production, where such materials play a role in contributing to environmental sustainability [6]. Lignocellulosic biomass consists mostly of polymer sugars (celluloses and hemicelluloses) and lignin [7,8]. It can be broken down into simple sugars by enzymatic hydrolysis or chemically by sulfuric or other acids [9]. Due to the process that requires less energy in mild conditions, enzymatic hydrolysis is becoming a more suitable pathway in biomass hydrolysis [10]. It is an important step in converting cellulose to glucose in pretreated biomass, which is carried out by cellulose enzymes in temperature range from 40 to 50 °C, with a pH range from 4 to 5 [11]. The degree of pretreated biomass, such as lignin removal, enzyme loading, and duration of hydrolysis is highly dependent on the enzymatic hydrolysis efficiency, since the process is also highly affected by cellulose crystalline structure [12]. The enhancement of the enzymatic hydrolysis process is possible by adding non-ionic surfactants, which can change the surface properties of cellulose, as well as reduce enzyme

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<https://www.mdpi.com/journal/molecules> Molecules 2021, 26, 753 2 of 23 loading. Such non-ionic surfactant is found to be polyethylene glycol (PEG), which can reportedly increase the convertability of lignocellulosic biomass for more than 30% [11,13,14]. Biofuels based on biomass have many advantages over fossil fuels: besides contributing to fuel diversity, different biofuels are accessible by different common biomass sources, have an environmentally friendly impact and potential, and provide many benefits in terms of economy and environment for all users of biofuels. Such biofuels are biodegradable and immensely contribute to sustainability. In addition, biofuels add value to migrating greenhouse gas (GHG) emissions, which provide a cleaner and more sustainable energy source with reduced air pollution. By using biomass feedstocks for bioethanol production, such actions of biomass usage enable the emerging development of rural areas in different countries, as well as increase of agricultural income. Such developing countries have more available land with favorable climate conditions and therefore minimum or at least lower labor costs. Another advantage with large-scale biofuel production for developing

Extended Abstract

countries is the reduction of its oil import dependence, which contributes to international competitiveness. When referring to the production of “good” bioethanol (bioethanol being the most commonly used biofuel for transportation worldwide) in terms of reducing the GHG emissions, it is important to replace polluting fossil fuels with more environmentally friendly lignocellulosic biomass. To ensure beneficial properties of biofuels, all kinds of by-products in the production process should be properly and efficiently utilized in order to minimize the GHG effect, as well as maximize their energy. In addition, emissions (such as carbon dioxide and nitrous oxide) should be kept to a minimum in terms of pollution and fertilizers, respectively. Moreover, biofuels such as bioethanol can help reduce the carbon dioxide escalation by replacing the fossil fuels and recycling the carbon dioxide being released when combusted as fuel [15]. However, in the ever-growing biofuel industry, sustainable energy systems and energy efficiency have an important decisive part, especially when renewable energy potentials compete with high energy demands.

Biography :

Dr. Bhatia working as a Post Doctorate Fellow in the Deptt. of Biotechnology Himachal Pradesh University Summer Hill, Shimla. He is working in the area of biofuel technology from last two year especially on the biotransformation lignocellulosic waste biomass into biofuels. He has eight years research and teaching experience in various areas of biotechnology. During his Doctoral research he developed bioprocess for the production of anticancer compounds and has published more than 20 research papers in various reputed journals, with more than 150 citations. He is also a budding bio- entrepreneur and acting as a team leader in his startup related to bio-transformation of solid waste into biofuel

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