



Effects of low intensity ultrasound on cellulase pretreatment

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ABSTRACT

This research was to explore the mechanism of ultrasonic impact on free cellulase activity and immobilize cellulase activities. The highest free cellulase activity was achieved when the sample was treated with low intensity ultrasound at 15 W, 24 kHz for 10 min, under which the enzyme activity was increased by 18.17% over the control. Fluorescence and CD spectra revealed that the ultrasonic treatment had increased the number of tryptophan on cellulase surface slightly, with the deformation of certain number of α -helix structure and increase of random coil content in cellulase protein. The highest immobilized cellulase activity was achieved when the sample was treated with low intensity ultrasound at 60 W, 24 kHz for 10 min, under which the enzyme activity was increased by 24.67% over the control. Scanning electron microscopy revealed that the ultrasonic treatment had increased the surface area of immobilized cellulase.

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1. Introduction

Bioethanol produced from lignocellulosic biomass is an interesting alternative source of energy since lignocellulosic raw materials do not compete with food crops and they are also less expensive than conventional agricultural feedstocks (Alviraa et al., 2010). To achieve an efficient conversion of waste cellulose to soluble sugars, the enzymatic hydrolysis of cellulose is suggested to be preferred to the various acid catalyzed processes using inorganic acid, subcritical or supercritical waters, etc. since the former not only offers a bioconversion process under the simpler and milder operating conditions but also produces no by-products detrimental to fermentative microorganisms (Li et al., 2005). Although the enzymatic route has the highest current cost, it has long-term potential for cost reduction (Zheng et al., 2009). Therefore, the present cellulosic ethanol research is driven by the need to reduce the production cost.

The bioethanol process based on the enzymatic hydrolysis of cellulose primarily includes three steps such as biomass pretreatment, enzymatic hydrolysis and fermentation, in which the hydrolysis step is one of the major contributors to the total production cost. Typically, it accounts for over 20% of the total production cost (Goh et al., 2010). A reduction in cellulase production cost, an

improvement in cellulase performance, and an increase in sugar yield are all vital to reduce the processing costs of bioethanol. Cellulases are relatively costly enzymes, and a significant reduction in cost will be important for their commercial use in bioethanol processing. Cellulase-based strategies that will make the bioethanol processing more economical include: increasing commercial enzyme volumetric productivity, producing enzymes using cheaper substrates, producing enzyme preparations with greater stability for specific processes, and producing cellulases with higher specific activity on solid substrates. It is also an important way to enhance the commercial cellulase activity by molecular modification (Singh et al., 2010).

Recently, application of ultrasonic technology in biological processing has widely attracted attention. Ultrasound has been used in the laboratory or processing plant to effect novel changes in the physicochemical properties of biological material in various areas, such as nanoemulsion preparation (Kentish et al., 2008), ultrasound-assisted extraction (Vilkhu et al., 2008), and reduction of viscosity (Iida et al., 2008). More recently the interest of food technologists has turned to the use of power ultrasound in altering enzyme activities. Prolonged exposure to high-intensity ultrasound has been shown to inhibit the catalytic activity of a number of enzymes (Kadkhodae and Povey, 2008). However, in some cases, enzyme activities have been found to have increased activity following short exposures to ultrasound (Duan et al., 2011; Lee et al., 2008). Some authors have recently reported that ultrasound can accelerate the enzymatic hydrolysis of solid leather waste (Jian et al., 2008). Little is known about the effects of ultrasonic

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