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Editorial

Genetic Engineering for Cellulosic Ethanol Production

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Introduction

Cellulosic ethanol production has attracted more and more interests due to depletion of fossil fuels as well as global warming. However, there are still some challenges to be addressed to realize its industrial production economically and environmentally friendly. One of the most important topics is construction of robust fermenting microorganisms which could tolerate inhibitors derived from pretreatment, secret cellulase and ferment both pentose and hexose simultaneously. Genetic modification could help us to obtain the required fermenting microorganisms and feedstock. Many works have been done on this aspect but there are still some works need to be carried out.

The Importance of Developing Cellulosic Ethanol And Its Main Technical Barriers

With the depletion of non-renewable fossil fuels and the increase of demand for fuels, utilization of low-cost renewable lignocellulosic biomass to produce fuel ethanol is thought to be a way to solve such problems [1]. Additionally, utilization of lignocellulosic biomass is recognized as a low-carbon process [2]. Under such conditions, development of cellulosic ethanol has been studied for many years. The whole process includes pretreatment of biomass, saccharification and fermentation, and distillation [3]. The technical barriers of pretreatment are lack of effective, low-cost, and environmentally friendly pretreatment methods, inhibitors generation during pretreatment, and detoxification of the resultant substrate. For saccharification and fermentation, barriers are low digestibility of polysaccharides and low convertibility of sugar into ethanol. Genetic engineering provides us a tool to address these problems.

Genetic Modification-Towards Fermenting Microorganisms

Genetic engineered microbes to tolerate or metabolize inhibitors generated from pretreatment

Inhibitors including furfural, 5-hydroxymethylfurfural, phenolic, and weak acids depress the growth and metabolism of microbes strongly and thus depress the fermentation performance [4]. Detoxification step is always essential for pretreated biomass especially for those pretreated with high severity. This step increases the overall cost of cellulosic ethanol production since additional equipment, chemicals, enzymes, and others may involved in this step. It has been known that furfural and 5-hydroxymethylfurfural could be metabolized by yeast [5], suggesting that genes related to conversion of such inhibitors exist in genome of yeast. Thus, using genetic engineering to enhance the convertibility of such inhibitors will release the inhibitory effect further. Laccase has been used widely to treat phenolics for efficient fermentation [6]. Expression of the genes encoding laccase in a selected fermenting microbe may be able to make it tolerate phenolics and ferment sugars into ethanol more efficiently. Using genetic engineering to breed robust fermenting microbes is an important direction.

Development of consolidated bioprocessing by using genetic engineering

Consolidated bioprocessing is thought to be a highly integrated configuration with lower cost and higher efficiency than the others with cellulase involvement [7]. There are two strategies for implementing consolidated bioprocessing. One is based on genetic modification of microorganisms with native cellulolytic ability and the other is based on genetic modification of those with good ethanol production performance [7]. Both strategies need genetic engineering to make the selected microorganism produce either ethanol from sugars or hydrolytic enzymes for hydrolyzing biomass. Many works have been conducted focusing on this aspect [8-10]. However, the saccharification and fermentation efficiency is not high enough to realize a scale-up production [11]. In addition, most of the studies use model substrates such as phosphoric acid swollen cellulose, reactive amorphous cellulose, and bacterial microcrystalline cellulose. Therefore, more studies are needed to explore how to use pretreated lignocellulosic biomass and enhance the efficiency by using consolidated bioprocessing.

Construction of co-fermenting microbes by using genetic engineering

Under such circumstances, genes encoding xylose reductase, xylitol dehydrogenase, and xylulokinase should be integrated into *Saccharomyces cerevisiae* by using genetic engineering [12]. These have been studied extensively. However, the convertibility of xylose into ethanol is not high enough which is caused by either byproducts formation or glucose inhibition. Furthermore, xylose fermentation of such recombinant yeast is more sensitive than glucose consumption to inhibitors including furfural, acetic acid, and phenol [13]. It is urgent to construct robust xylose-fermenting strains by using genetic engineering.

Genetic Modification-Towards Lignocellulosic Feedstock

To avoid complex pretreatment procedures and lower the pretreatment severity, composition and structure of lignocellulosic biomass are modified by genetic engineering [14]. Through genetic modification, the resultant biomass could be hydrolyzed more easily after a mild pretreatment process. This decreases the formation of inhibitors and the cost of saccharification. Therefore, genetic modification of plant for bioethanol production is also a hot topic.

In conclusion, to address the bottlenecks of bioethanol production from lignocellulosic materials by using genetically engineered fermenting microorganisms or feedstock is promising.

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