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Genetic engineering approaches for enhanced lignocellulosic-based bioprocesses

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Lignocellulosic biomass is the most abundant, low-cost, bio-renewable resource. It has a recognised potential as a sustainable platform for the production of biofuels and other bio-chemicals. To improve the accessibility of the cellulose component from complex lignocellulosic structures to the enzymes, a pretreatment step is necessary. Enzymatic saccharification of resulting whole slurry is highly desirable as it avoids the solid-liquid separation step, the need for detoxification and related waste disposal problem, and increases final sugar concentration. However, lignin residues and other inhibitory compounds resulting from pretreatment negatively affect the digestibility of the whole slurry and compromise fermentation efficiency. To tackle these pitfalls, genetic engineering strategies have been developed and integrated in the process to improve both stages.
For improving the fermentation efficiency, our approach has been to intensify the process by using high solid loadings and both pentose and hexoses fractions, enriching sugar concentration available for fermentation. To work under such demanding conditions robust yeast strains are crucial. We have selected natural robust yeast isolates and identified key genes necessary for yeast growth and maximal fermentation rate in hydrolysates. Selected robust yeast chassis have been metabolic engineered for co-fermentation of glucose and xylose from hemicellulose fraction using a novel metabolic assembly tool and key tolerance genes expression has been simultaneously evaluated for the valorization of biomass of different origins. Results obtained pointed to the importance of designing from the very beginning a tailor-made yeast considering the specific raw material and process [1]. The flexibility of the metabolic assembly tool developed and the selected robust yeast backgrounds envisioned the developing of effective yeast platforms for biomass processing into different products. For improving the saccharification of whole slurry, our strategy has been to use the efficient recombinant protein production system from *Escherichia coli* to produce hydrolysis enhancers, namely a family 3 carbohydrate-binding module (CBM3). The purified CBM3 was used as an additive in the enzymatic hydrolysis of the whole slurry from hydrothermally-pretreated *Eucalyptus globulus* wood among other biotechnological applications [2]. The results obtained show an increase in glucose yield when CBM3 was added, compensating the negative effect of inhibitors on the enzymatic efficiency of whole slurry saccharification. Thus, CBM3 is a valid additive for enhanced lignocellulosics saccharification and a valuable alternative to costly additives (e.g. BSA) as it can be affordably obtained from heterologous bacterium or integrated in the developed yeast platforms, thus contributing to more cost-efficient and environmental-friendly biomass conversion bioprocesses.

References

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