# Abstract

Bioethanol from lignocellulosic feedstocks is an attractive renewable energy source to reduce world's dependence on depleting petroleum-derived non-renewable energy sources. However, large-scale microbial production of bioethanol from lignocelluloses faces several challenges and one major challenge is the absence of microorganisms that can efficiently ferment all sugars released during hydrolysis of lignocelluloses. Genetic engineering of traditional strains and the utilization of specialized microbial communities have been proposed as two practical approaches to overcome this problem.

Genome-scale metabolic network, a database of all the available information of an organism's metabolism, is now reconstructed for many useful microorganisms and is viewed as an effective tool for metabolic phenotype prediction. Dynamic flux balance analysis (dFBA) is an efficient constraint-based approach to analyse such complex networks under unsteady state situations in batch/fed-batch bioreactors. Validated genome-scale metabolic models that are capable of making quantitative predictions are very important tools for understanding and engineering the behaviour of microorganisms.

This research develops and validates genome-scale dFBA models for various mono-culture and co-culture systems of specialized microbes and performs *in silico* analysis and optimization of bioethanol production from glucose/xylose mixtures. In particular, this study investigates dFBA models for microorganisms such as *Saccharomyces cerevisiae*, *Escherichia coli*, *Scheffersomyces stipitis*, and *Zymomonas mobilis* and presents a detail *in silico* analysis of their bioethanol production potential. The enhancement in bioethanol production is investigated both in terms of process systems engineering approach and *in silico* genetic engineering approach. The identification of appropriate genetic manipulations to be applied to an organism for desired metabolite production is an important task for performing metabolic experiments. The predictions of our *in silico* genetic engineering studies will give valuable guidance for reducing the number of such complex and time consuming *in vivo* experimental trials.



**Keywords:** ***Bioethanol***, ***Co-culture***, ***Dynamic flux balance analysis***, ***Genome-scale metabolic network***, ***Mono-culture***, ***Optimization***