Screening and genetic improvement of microorganisms for production of fuels and chemicals

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Lignocellulosic biomass, which is rich in pentose and hexose sugars, and crude-glycerol waste of biodiesel industry, are two inexpensive and abundant substrates that can be used for production of fuels and chemicals. In this context, there is an increasing interest in identify and develop microbial strains able to efficiently convert these substrates to higher value-added products. In this work, yeasts and bacteria from different habitats have been isolated, prospected and genetically improved to produce fuels and chemicals from xylose and crude-glycerol. Initially, approximately 500 yeast and 5000 bacterial strains were screened for growth in micro-titer plates containing either xylose or glycerol as the only carbon source. Afterwards, tolerance of selected strains towards contaminants of the substrates was also evaluated for growth in sugarcane bagasse hydrolysate and crude glycerol from a biodiesel industry. Based on these results, we were able to identify yeast and bacterial strains for evaluation under fermentative conditions. In parallel, sensitivity of known xylose-fermenting yeast strains to lignocellulosic hydrolysate inhibitors was systematically evaluated. Afterwards, three strains were genetically improved for fermentation of lignocellulosic hydrolysates by evolutionary engineering. Analysis of substrate consumption and product formation demonstrated that the newly isolated and the genetically improved strains can efficiently convert xylose and/or glycerol to a variety of chemicals, for example, 2,3-butanediol, 1,3-propanodiol, xylitol and ethanol. Our results demonstrated the potential of bioprospection and evolutionary engineering to obtain new yeast strains for biotechnological applications.