An NGS approach to analysing HMF resistance in Saccharomyces cerevisiae

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Abstract

Bioethanol is the most promising renewable fuel to substitute fossil fuel and it is generated as a product of fermentation of sugars, which can occur throught two processes, first generation (1G) and second generation (2G). They differ basically in the raw material used, where, in brazilian production, 1G require sugarcane juice, while 2G, unused plant parts (bagasse), which are rich in polymers such as lignin, cellulose and hemicellulose. Both methodologies involve subproducts that interfere in the metabolism of microorganisms used to fermentation step, being Saccharomyces cerevisiae the most commonly used. While first generation ethanol exhibits factors such as temperature, O2 pressure, pH, alcohol concentration and contaminants as inhibitors, second generation, for requesting a thermic preprocessing to expose the fibres, and to convert cellulose and hemicellulose to simple monomers, shows, besides 1G inhibitors, acetic acid, furfural and HMF - hydroxymethylfurfural. In summary it introduces the importance of understanding yeast's resistance to increase productivity. In this work we studied a diploid industrial strain resistant to HMF that was sporulated, beeing selected four resistent spores and three non resistent spores. These eight strains were sequenced using Illumina paired-end technology and the data was analysed. The pipeline includes quality analysis of the reads (FastQC), genome assembly of a non resistant strain (SPAdes), gene prediction (Augustus), variant calling (GATK) and effect prediction (VEP), gene annotation (Blastn), chromosomal mapping (MUMmer package) and gene ontology classification (SGD website). This pipeline allowed us to identify a set of 68 candidate genes that could be related to HMF robustness. Future perspectives include the application of this pipeline to a greater set of sequenced strains, structural analysis of the proteins that are translated from the found genes, and experimental validation in order to determine a mechanism compatible with the resistance under study.

Funding: Asociación de Universidades Grupo Montevideo (AUGM), FAPESP