

RNAi Assisted Genome Evolution Unveils Yeast Mutants with Improved Xylose Utilization

Background/objective

Lack of an efficient means of fermenting xylose to ethanol has hampered full valorization of abundant lignocellulosic feedstocks. Conventional bioengineering methods were ineffective at further enhancing xylose uptake and fermentation in the xylose-consuming mutant *Saccharomyces cerevisiae* SR8. Researchers sought to overcome this barrier using the novel RNAi Assisted Genome Evolution (RAGE) technique.

Approach

- ❖ *S. cerevisiae* SR8 was engineered via three rounds of RAGE. Gene overexpression and down-regulation were estimated via reverse-transcription followed by qPCR.

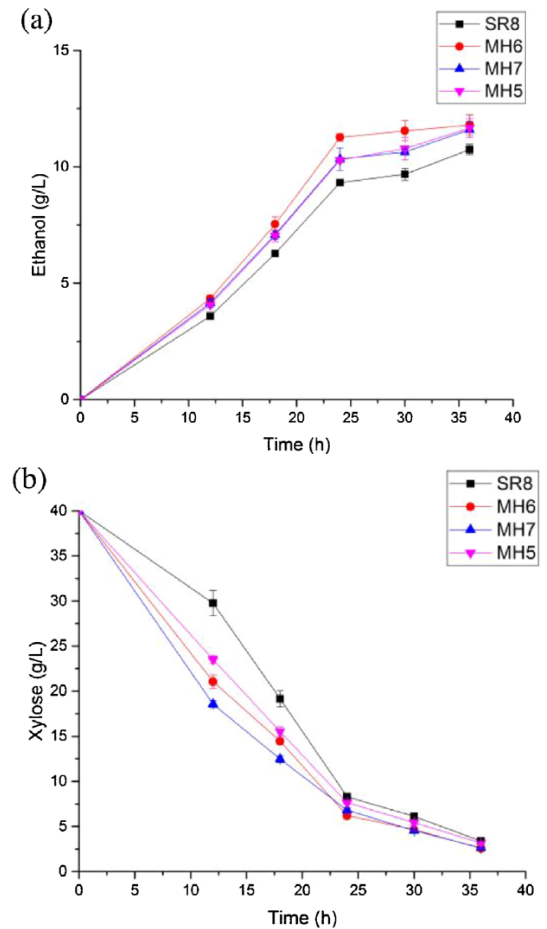
Results

- ❖ Up-regulation of the protein *VPS13* and down-regulation of *CDC11* were found to optimize xylose utilization.
- ❖ After two rounds of RAGE, xylose utilization rate and ethanol productivity were improved by 29% and 45%, respectively. An additional round of RAGE did not yield further improvements.

Significance

- ❖ The improved *S. cerevisiae* SR8 strain with substantially enhanced xylose fermentation capability will increase ethanol production capacity from lignocellulosic biomass.
- ❖ RAGE successfully increased the efficiency of an important engineered bioprocess where other conventional bioengineering methods had failed, suggesting that RAGE may be a useful tool for enhancing the production of other valuable bioproducts from yeasts.

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Xylose utilization (a) and ethanol production (b) in SR8 and all mutants identified in the second round of RAGE