CRISPR-Cas9 tool expedites production of biofuel precursors and specialty polymers in living systems

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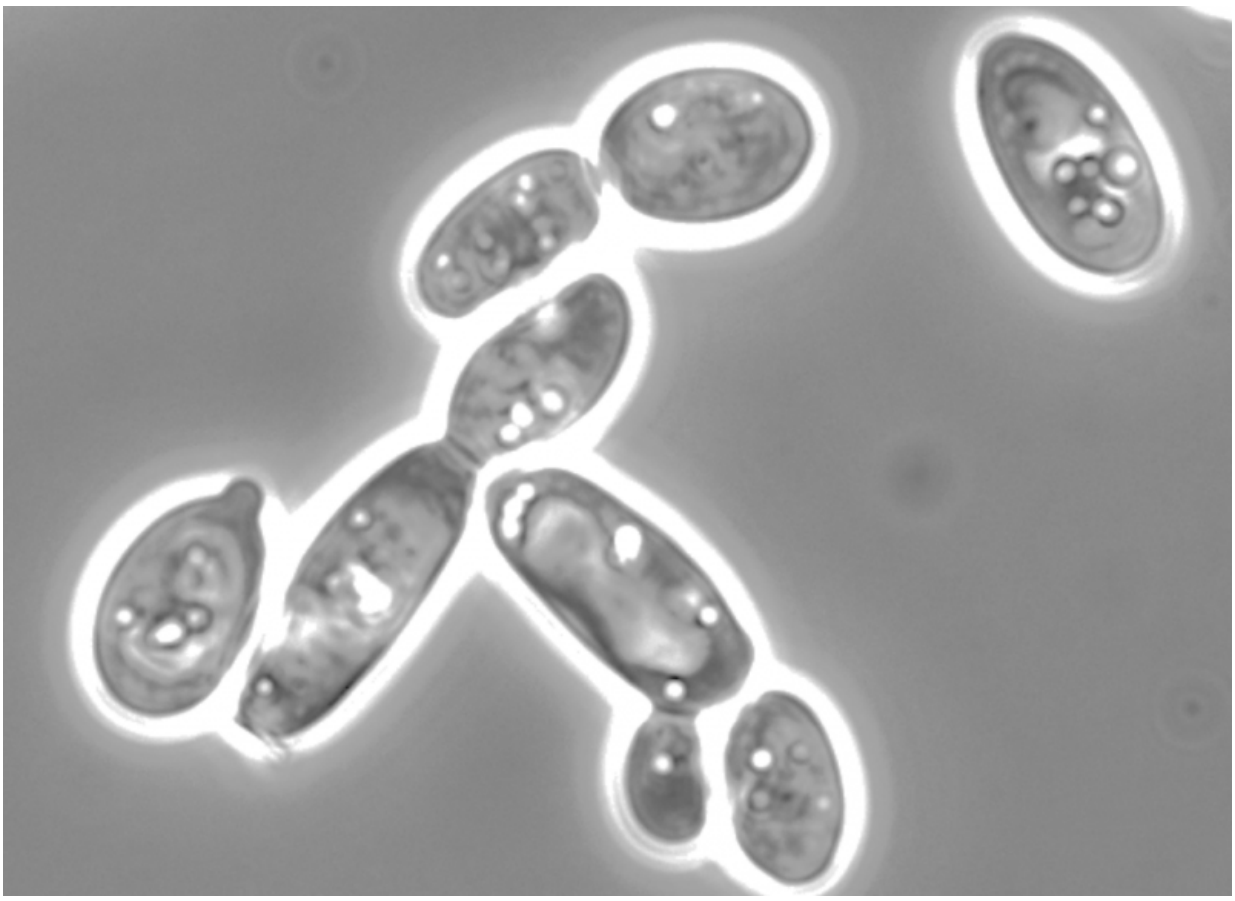

Bright-field microscopy of *Yarrowia lipolytica*. Gene editing in this yeast strain will lead to new precursors for biofuels and specialty polymers. Credit: University of California, Riverside

A team led by a researcher at the University of California, Riverside has adapted the CRISPR-Cas9 gene editing system for use in a yeast strain that can produce useful lipids and polymers.

The development will lead to new precursors for biofuels, specialty polymers, adhesives and fragrances.

Published recently in the journal *ACS Synthetic Biology*, the research involves the oleaginous (oil-producing) yeast Yarrowia lipolytica, which is known for converting sugars to lipids and hydrocarbons that are difficult to make synthetically. Until now, Y. lipolytica has been hard to manipulate at the genetic level, but the application of CRISPR-Cas9 will change that, allowing scientists to tap into its bio-manufacturing potential.

Described in 2012, CRISPR-Cas9 is a groundbreaking technique that enables scientists to make precise targeted changes in living cells. Unlike traditional gene-editing methods, it is cheap, easy to use and effective in almost any organism.

"Traditionally, researchers have focused on model organisms that are relatively easy to manipulate at the genetic level, and those working on less tractable species have had to go through long and tedious processes to create new strains. Our work with Y. lipolytica is a good example of how the CRISPR-Cas9 system is facilitating research in organisms that are biologically interesting but historically difficult to work with," said Ian Wheeldon, an assistant professor of chemical and environmental engineering at UCR's Bourns College of Engineering and the study's principal investigator.

In the paper, the team adapted CRISPR-Cas9 for Y. lipolytica, showing that the system could be used to knock genes out and introduce new genes, both useful tools in bio-manufacturing.

Wheeldon said the current work was the first step in a National Science Foundation-funded project to create long chain hydrocarbons—used to make specialty polymers, adhesives, coatings and fragrances—from yeast rather than synthetically.

"Currently, these molecules are produced from non-renewable raw materials derived from petroleum in processes that are inefficient and pose safety risks, so being able to produce them from cheap raw materials in a bio-manufacturing process is very appealing," Wheeldon said.

Other researchers may use the system to create precursors for biofuels, reducing the current reliance on edible plant oils, Wheeldon said.

More information: Cory M. Schwartz et al. Synthetic RNA Polymerase III Promoters Facilitate High-Efficiency CRISPR–Cas9-Mediated Genome Editing in , *ACS Synthetic Biology* (2016). DOI: 10.1021/acssynbio.5b00162

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