New discovery opens novel pathway for high-titer production of drop-in biofuels

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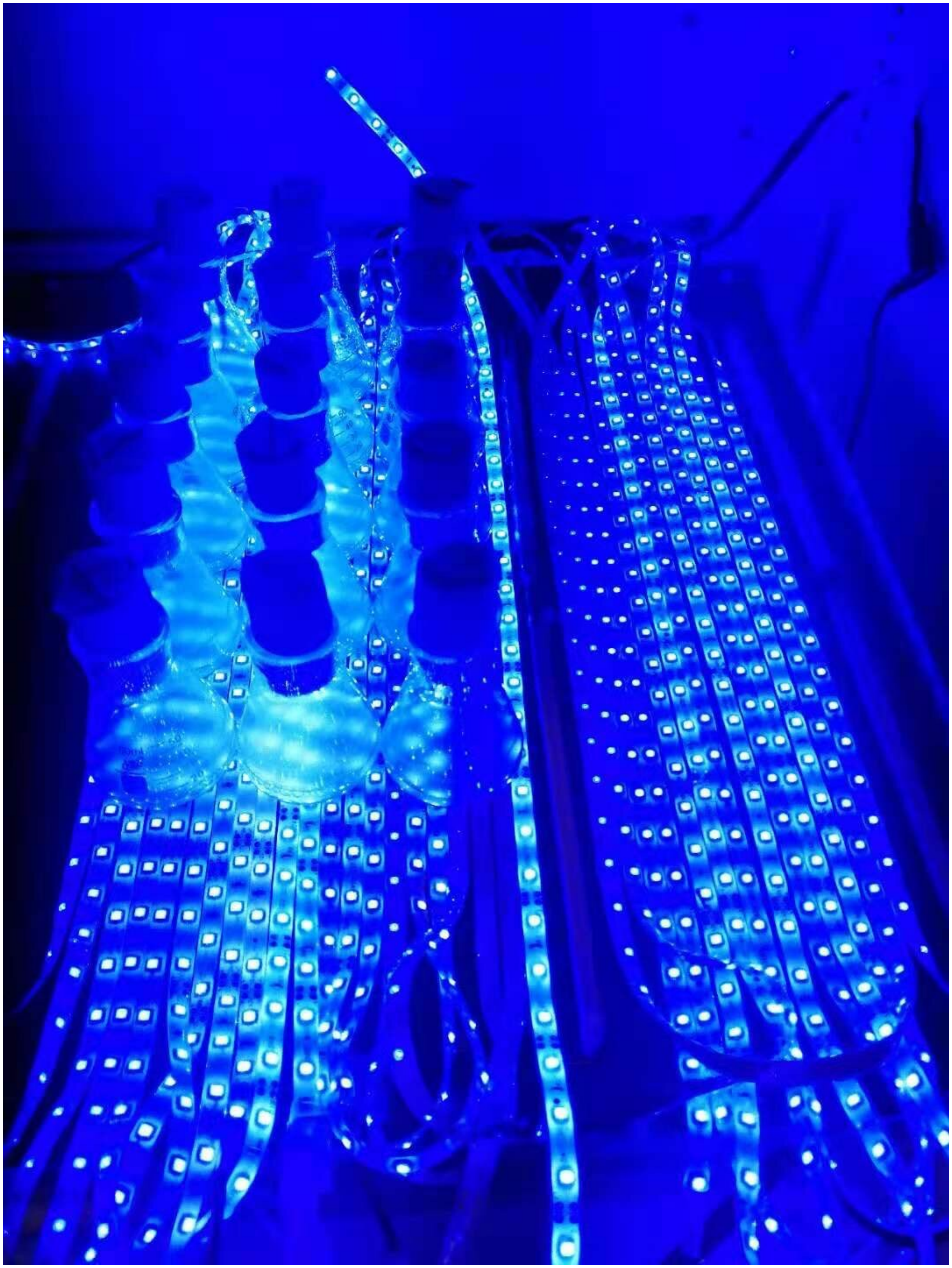

The lab-setup of the light incubator showing two different experimental blue light setups. Credit: Jingbo Li, MIT

A special light-dependent enzyme, which was first discovered about three years ago, is the focal point in a new scientific discovery that enables high-yield production of drop-in biofuels from biomass.

In a study now published in *Nature Communications*, engineers from Aarhus University and Massachusetts Institute of Technology have proved, that the original assumption of the enzymatic process in this biomass-to-biofuels conversion is actually wrong.

The findings have allowed the researchers to successfully biosynthesize green fuels at close to industrially relevant levels of 1.47 gram per liter from glucose.

The light-dependent enzyme, which originates from microalgae, has the particular characteristic that it can decarboxylate fatty acids into alkanes (thus converting cellulosic biomass into drop-in biofuels) using blue light as the only source of energy.

The researchers artificially insert the enzyme into the cells of the oleaginous yeast Yarrowia Lipolytica thereby engineering its metabolism. The yeast synthesizes glucose, originating from biomass, into lipids (specifically the molecules free fatty acids and fatty acyl-CoAs) which is then converted to alkanes by the enzyme in a metabolic reaction called fatty acid photodecarboxylase—in short FAP.

But ever since the discovery of the enzyme, it has been assumed, that free fatty acids are the enzyme's preferred reactant in the FAP process; that an abundance of free fatty acids would result in higher yield biofuel production.

This assumption is wrong, however.


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The lab-setup of the light incubator showing two different experimental blue light setups. Credit: Jingbo Li, MIT

"In our study, we have proved that fatty acyl-CoA—and not free fatty acid—is the preferred reactant for the light-dependent enzyme. This finding has been successfully used in our study to metabolize 89 percent of fatty acyl-CoA into alkanes, reaching titers of 1.47 g/l from glucose," says Bekir Engin Eser, an assistant professor at Aarhus University.

The predominant production of oleochemical based drop-in fuels today are made by converting 'conventional' oleochemicals such as vegetable oils, used cooking oils, tallow, and other lipids to hydrocarbons (mainly alkanes) using energy intense chemical treatment methods.

However, sourcing large quantities of more or less sustainable lipid feedstocks at a low enough cost to result in profitable drop-in biofuel production remains a challenge that severely limits the expansion of this production platform. And furthermore, this production is competing with food supply.

Biosynthesis constitutes a cheap and sustainable solution, where the production is instead based on the conversion of cellulosic biomass—the most abundant renewable natural biological resource available on Earth.

Biological synthesis of alkanes from fatty acids is not a native, preferable metabolic pathway for the yeast however, since alkanes are toxic to its cells. Therefore, researchers use special ability enzymes for this purpose and encode the corresponding genes into the cells of the yeast.

The new discovery is a possible breakthrough in biosynthesis of drop-in fuels, since the researchers—for the first time ever using this process—have utilized the new knowledge to synthesize green fuels at a level that's relevant for future industrial production:

"Previous metabolic engineering studies would target maximizing the concentration of free fatty acids in the cells that are being engineered. But now, with this discovery, we know that it is fatty acyl-CoA that needs to be maximized. This is important news for synthetic biology applications, and we can now begin to maximize the flux of the fatty acyl-coA into this engineered metabolic pathway to reach even higher titers in the future," says Associate Professor Zheng Guo from Aarhus University.

More information: Jingbo Li et al, Synthesis of high-titer alka(e)nes in Yarrowia lipolytica is enabled by a discovered mechanism, *Nature Communications* (2020). DOI: 10.1038/s41467-020-19995-0

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