

Four strains of bacteria work together to produce pigment for food and cosmetics industry

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Credit: Rensselaer Polytechnic Institute

Nature makes the vibrant pigments manufacturers want for foods and cosmetics, but getting them from plants to the products we buy is so difficult that many manufacturers rely on artificial colors. Now, researchers at Rensselaer Polytechnic Institute have shown that four strains of *E. coli* bacteria working together can convert sugar into the natural red anthocyanin pigment found in strawberries, opening the door to economical natural colors for industrial applications.

The research marks the first biosynthesis method using four [strains](#) of [bacteria](#) to manufacture a compound in a single step, said Mattheos Koffas, a professor of chemical and biological engineering at Rensselaer, and member of the Center for Biotechnology and Interdisciplinary Studies. Results appear in *mBio*, a publication of the American Society for Microbiology.

"For the first time, we are able to completely synthesize anthocyanins in a biological system," said Koffas. "We feed the bacteria glucose and they do the rest. This demonstrates that an inexpensive technology can produce these valuable compounds."

Manufacturers are increasingly interested in natural colors, a shift founded in research documenting the health hazards of artificial colors for foods and cosmetics. Natural pigments found in [plants](#) such as anthocyanins, carotenoids, or lycopene produce indisputably safe colors. Anthocyanins exist in a far broader spectrum than other pigment classes

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producing all colors except green—and are responsible for natural [color](#) found in foods like blueberries, raspberries, black rice, and the hues of autumn leaves.

But producing anthocyanins for industrial use is challenging: plant-derived pigments require costly processing and produce inconsistent

results; and because [anthocyanin](#) molecules are complex, chemists have been unable to synthesize them.

Koffas' lab has been investigating a production method of anthocyanins involving genetic engineering since 2005. Sixteen genes govern the production of anthocyanin in plants. But transferring those genes, and the molecular pathway they trigger, from a plant to a bacteria is not a straightforward process, said J. Andrew Jones, first author of the paper and a doctoral student at the time the research was conducted. To adapt the molecular pathway from a plant to the bacteria, the Koffas lab pieced together genes from bacteria, yeast, and six different varieties of plants.

In early research, the Koffas lab introduced the entire molecular pathway it had assembled into a single strain of E.coli bacteria. But the pathway proved too long for the cell, which tended to use the additional genes to produce its own metabolites instead of anthocyanins, said Jones, currently a member of the faculty at Miami University of Ohio.

The current research divides the entire pathway among four different strains of bacteria, modified to assemble anthocyanin in stages. Researchers divided the [pathway](#) into "modules" that produced intermediates that easily diffuse out of the bacterial cell. When combined in a single flask, the first bacteria ingest sugar and produce "intermediate" compounds, phenylpropanoic acids, which are ingested by the second bacteria, which produces a second intermediate, and so forth until the fourth strain produces anthocyanin.

In the next stage of work, researchers will optimize each stage of the process. Each of the four strains was chosen based on its ability to produce its assigned intermediate; however, some segments produce more than others, and the final output is modest, producing milligrams per liter of anthocyanin. Production will have to approach of hundreds

of milligrams per liter to be a commercially viable approach.

"I have no doubt that production of anthocyanins from a recombinant microbial host is the only viable method for making these compounds in an economically sustainable manner," said Koffas.

More information: J. Andrew Jones et al. Complete Biosynthesis of Anthocyanins Using *E. coli* Polycultures, *mBio* (2017). [DOI: 10.1128/mBio.00621-17](https://doi.org/10.1128/mBio.00621-17)

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