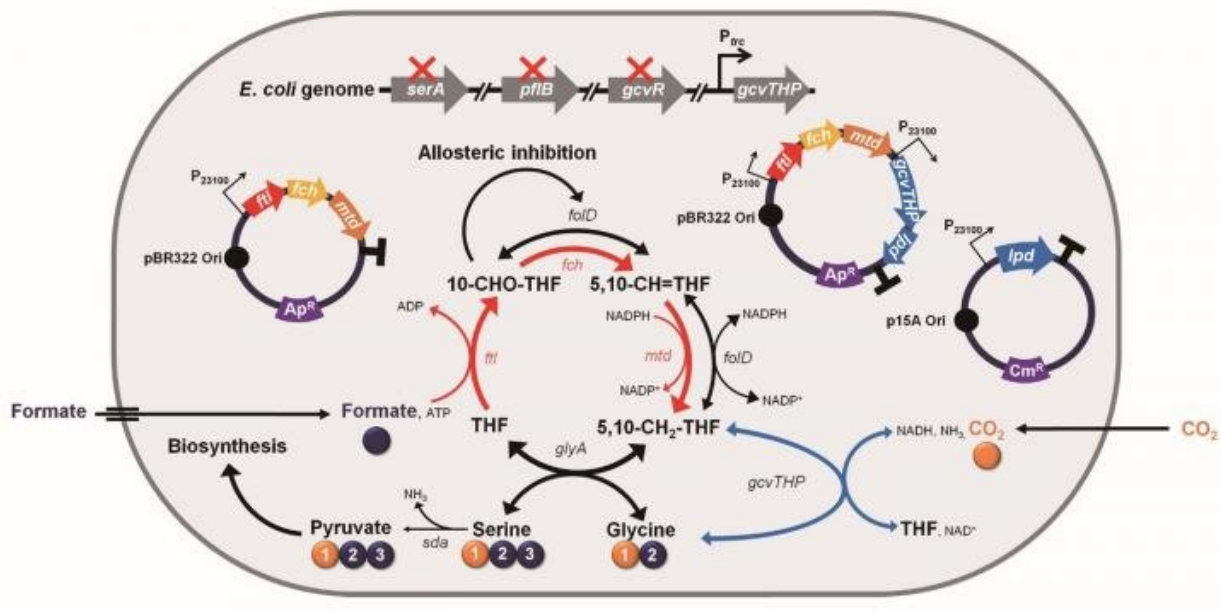


# Engineered E. coli using formic acid and CO<sub>2</sub> as a C1-refinery platform strain

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Formic acid and CO<sub>2</sub> assimilation pathways consisting of the reconstructed THF cycle and reverse glycine cleavage reaction. This schematic diagram shows the formic acid and CO<sub>2</sub> assimilation procedure through the pathway. Plasmids used in this study and the genetic engineering performed in this study are illustrated. Credit: KAIST

A research group at KAIST has developed an engineered *E. coli* strain that converts formic acid and CO<sub>2</sub> to pyruvate and produces cellular energy from formic acid through reconstructed one-carbon pathways.

The strategy described in this study provides a new platform for producing value-added chemicals from one-carbon sources.

Formic [acid](#) is a carboxylic acid composed of one carbon. Formic acid was produced from CO<sub>2</sub> by the chemical method. Recently, the C1 Gas Refinery R&D Center has successfully developed a biological process that produces [formic acid](#) from carbon monoxide for the first time. Formic acid is in a liquid state when at room temperature and atmospheric pressure. In addition, it is chemically stable and less toxic, thus, easy to store and transport. Therefore, it can be used as an alternative carbon source in the microbial fermentation process. In order to produce value-added chemicals using formic acid, a metabolic pathway that converts formic acid into cellular molecules composed of multiple carbons is required. However, a metabolic pathway that can efficiently convert formic acid into cellular molecules has not been developed. This acted as an obstacle for the production of value-added chemicals using formic acid

A research group of Ph.D. student Junho Bang and Distinguished Professor Sang Yup Lee of the Department of Chemical and Biomolecular Engineering at the Korea Advanced Institute of Science and Technology (KAIST) addressed this issue. This study, titled "Assimilation of Formic Acid and CO<sub>2</sub> by Engineered Escherichia coli Equipped with Reconstructed One-Carbon Assimilation Pathways," has been published online in the *Proceedings of the National Academy of Sciences* on September 18.

There has been increasing interest in utilizing formic acid as an alternative carbon source for the production of value-added chemicals. This research reports the development of an engineered E. coli strain that can convert formic acid and CO<sub>2</sub> to [pyruvate](#) and produce cellular energy from formic acid through the reconstructed one-carbon pathways.

The metabolic pathway that efficiently converts formic acid and CO<sub>2</sub> into pyruvate was constructed by the combined use of the tetrahydrofolate cycle and reverse glycine cleavage reaction. The tetrahydrofolate cycle was reconstructed by utilizing *Methylobacterium extorquens* formate-THF ligase, methenyl-THF cyclohydrolase, and methylene-THF dehydrogenase. The glycine cleavage reaction was reversed by knocking out the repressor gene (*gcvR*) and overexpressing the *gcvTHP* genes that encode enzymes related with the glycine cleavage reaction. Formic acid and CO<sub>2</sub> conversion to pyruvate was increased via metabolic engineering of the *E. coli* strain equipped with the one-carbon assimilation pathway.

In addition, in order to reduce glucose consumption and increase formic acid consumption, *Candida boidinii* formate dehydrogenase was additionally introduced to construct a cellular energy producing pathway from formic acid. This reduces glucose consumption and increases formic acid consumption.

The reconstructed one-carbon pathways can supply cellular molecules and cellular energies from the formic acid and CO<sub>2</sub>. Thus, the engineered *E. coli* strain equipped with the formic acid and CO<sub>2</sub> assimilation pathway and cellular energy producing pathway from formic acid showed cell growth from formic acid and CO<sub>2</sub> without glucose. Cell growth was monitored and <sup>13</sup>C isotope analysis was performed to confirm *E. coli* growth from the formic acid and CO<sub>2</sub>. It was found that the engineered *E. coli* strain sustained cell growth from the formic acid and CO<sub>2</sub> without glucose.

Professor Lee said, "To construct the C1-refinery system, a platform strain that can convert one-carbon materials to higher carbon materials needs to be developed. In this report, a one-carbon pathway that can efficiently convert formic acid and CO<sub>2</sub> to pyruvate was developed and a [cellular energy](#) producing [pathway](#) from formic acid was introduced.

This resulted in an engineered E. coli strain that can efficiently utilize formic acid as a carbon source while glucose consumption was reduced. The reconstructed one-carbon pathways in this research will be useful for the construction of the C1-refinery system."

**More information:** Junho Bang et al, Assimilation of formic acid and CO<sub>2</sub> by engineered Escherichia coli equipped with reconstructed one-carbon assimilation pathways, *Proceedings of the National Academy of Sciences* (2018). [DOI: 10.1073/pnas.1810386115](https://doi.org/10.1073/pnas.1810386115)

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