

# Highly efficient production of diverse rare ginsenosides using combinatorial biotechnology

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## Abstract

The rare ginsenosides are recognized as the functionalized molecules after oral administration of *Panax ginseng* and its products. The sources of rare ginsenosides are extremely limited because of low ginsenoside contents in wild plants, hindering their application in functional foods and drugs. We developed an effective combinatorial biotechnology approach including tissue culture, immobilization, and hydrolyzation methods. Rh2 and nine other rare ginsenosides were produced by MeJA-induced culture of adventitious roots in a 10 L bioreactor associated with enzymatic hydrolysis using six  $\beta$ -glycosidases and their combination with yields ranging from 5.54-32.66 mg L<sup>-1</sup>. The yield of Rh2 was furthermore increased 7% by using immobilized BglPm and Bgp1 in optimized pH and temperature condition, with the highest yield reaching 51.17 mg L<sup>-1</sup> (17.06% of PPD-type ginsenosides mixture). Our combinatorial biotechnology method provides a highly efficient approach to acquiring diverse rare ginsenosides, replacing direct extraction from *Panax* plants, and can also be used to supplement yeast cell factories.

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