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Engineering of Heterologous Mevalonate Pathway in *Escherichia coli* for Enhanced Supply of Isoprenoids Precursors

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Extended Abstract

Isopentenyl diphosphate (IPP) is an important building block for Isoprenoids, which are the compounds of the most abundant natural secondary metabolite, such as isoprene, carotenoids, and terpenoids. Most prokaryotes synthesize IPP with 2-C-methyl-D-erythritol 4-phosphate (MEP) pathway, while most eukaryotes and some Gram-positive strains synthesize with mevalonate (MVA) pathway. Here we report cloning and reconstruction of six mevalonate (MVA) pathway genes of novel bacteria *Enterococcus Kingsejongenesis* which was isolated from a South Polar Brown Skua. Each gene encoding the six mevalonate (MVA) pathway enzymes was reconstructed as an individual synthetic expression module, and then was functionally expressed as a single module or assembled module in *Escherichia coli*. The recombinant *E. coli* expressing the upper assembled MVA pathway module (HMG_CoA synthase(mvaS) and Thiolase/HMG_CoA reductase(mvaE)) produced mevalonate up to 57g/L by fed-batch fermentation. For balanced and fine-turned protein expression in metabolic pathway, expression levels of mvaS and mvaE were regulated by synthetic expression cassette(SEC) library. Randomizing promoter strength and ribosome binding site (RBS), simultaneously, was employed to diversify expression of mvaS and mvaE protein specifically in *E.coli*. The *E.coli* strain engineered in this study can serve as the basis for creating an alternative way for production of mevalonate(mevalonolactone), which is industrially valuable and critical intermediate in isoprenoids biosynthesis pathway in diverse species, in place of production from chemical synthesis.