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Production of amino acids – Genetic and metabolic engineering approaches

Jin-Ho Lee [a], Volker F. Wendisch [b,*]

[a] Major in Food Science & Biotechnology, School of Food Biotechnology & Nutrition, Kyungsung University, 309, Suyeong-ro, Nam-gu, Busan 48434, Republic of Korea
[b] Genetics of Prokaryotes, Faculty of Biology and Center for Biotechnology, Bielefeld University, Bielefeld, Germany

HIGHLIGHTS

- Amino acid biosynthesis and transcriptional regulators and regulons reviewed.
- Rational metabolic engineering based on modulation of transcriptional regulators.
- Improved amino acid producing strains by genetic regulatory engineering.
- Functional tools improve systems metabolic engineering of amino acid producers.

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ABSTRACT

The biotechnological production of amino acids occurs at the million-ton scale and annually about 6 million tons of L-glutamate and L-lysine are produced by *Escherichia coli* and *Corynebacterium glutamicum* strains. L-glutamate and L-lysine production from starch hydrolysates and molasses is very efficient and access to alternative carbon sources and new products has been enabled by metabolic engineering. This review focusses on genetic and metabolic engineering of amino acid producing strains. In particular, rational approaches involving modulation of transcriptional regulators, regulons, and attenuators will be discussed. To address current limitations of metabolic engineering, this article gives insights on recent systems metabolic engineering approaches based on functional tools and method such as genome reduction, amino acid sensors based on transcriptional regulators and riboswitches, CRISPR interference, small regulatory RNAs, DNA scaffolding, and optogenetic control, and discusses future prospects.

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* Corresponding author.
E-mail address: volker.wendisch@uni-bielefeld.de (V.F. Wendisch).

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1. Introduction

Amino acids are predominantly used as food and animal feed additives as well as pharmaceuticals and cosmetics (Wendisch, 2014). Their demand has been steadily increased with annual growth rates of 5–7%. In 2014, the chemical production of D,L-methionine amounted to 1 million tons (http://corporate.evonik.com/_layouts/websites/internet/downloadcenterfilehandler.ashx?fileid=2551; accessed March-02-2017). In 2015, 3.1 million tons of L-glutamate and 2.4 million tons of L-lysine were produced by fermentation worldwide (http://www.ajinomoto.com/en/ir/pdf/Food-Oct2016.pdf; and /Life_Support-Oct2016.pdf; accessed March-02-2017). In addition, a wealth of chemicals with diverse applications has been generated from amino acids and precursors by microbial fermentation (Hirasawa and Shimizu, 2016; Jensen et al., 2015; Wendisch, 2014). Strains have been engineered for the conversion of alternative carbon sources including allowing for sustainable amino acid bioprocesses starting from e.g. lignocellulosics or agro-industrial residues (Wendisch et al., 2016).

Development of amino acid producing strains using mainly *Corynebacterium glutamicum* and *Escherichia coli* as hosts has been extensively achieved by targeted metabolic engineering along with classical mutagenesis and selection (Park and Lee, 2008; Wendisch, 2014) (Fig. 1). Despite all elaborate efforts, the target-specific engineering approach might have limitations in development of production strains with increased yield and productivity because engineered target genes were mainly confined to local metabolic pathways rather than global metabolic and regulatory networks. Recently, systems metabolic engineering that integrates metabolic engineering with systems and synthetic biology allowed successful

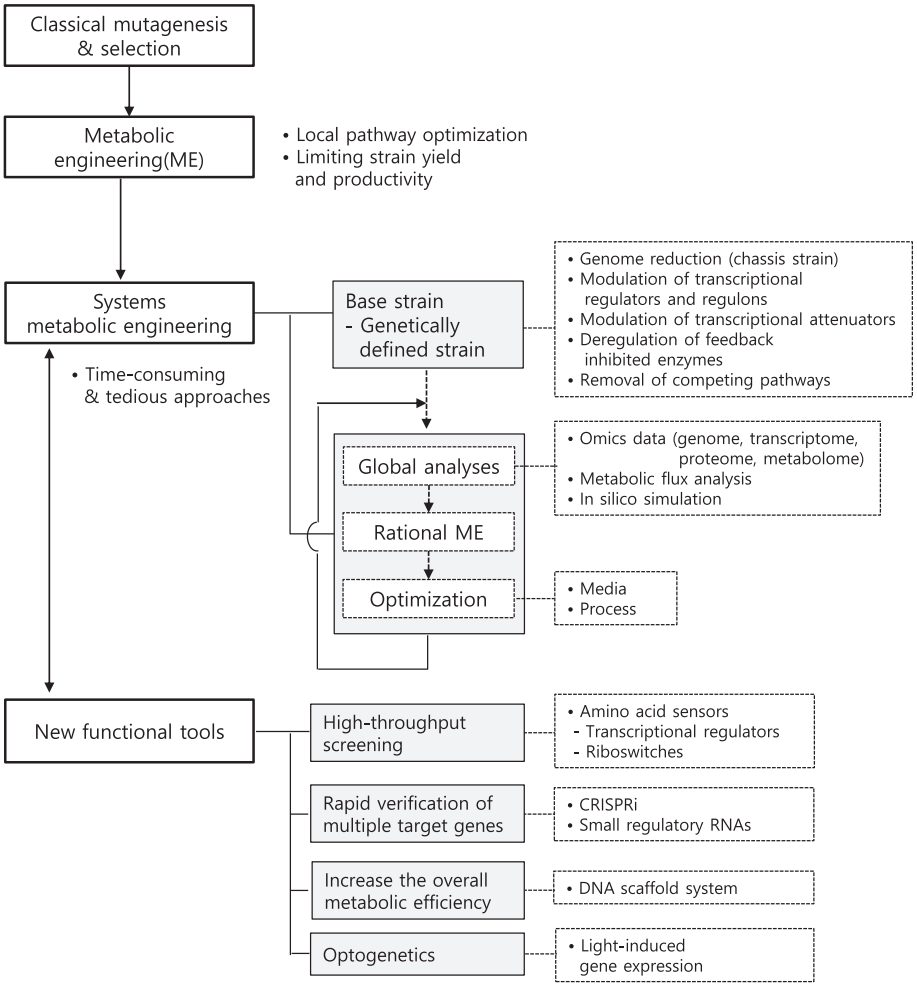

Fig. 1. Overall strain development procedure and related new functional tools for the production of amino acids.

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