CORYNEBACTERIUM GLUTAMICUM AS A PLATFORM STRAIN FOR THE PRODUCTION OF A BROAD VARIETY OF TERPENOIDS

Petra Peters-Wendisch, Bielefeld University, Germany
petra.peters-wendisch@uni-bielefeld.de
Arthur Burgardt, Bielefeld University, Germany
Sabine Selsemeyer-Voigt, Bielefeld University, Germany
Jonas Frohwitter, Hochschule Biberach, Germany
Nadja A. Henke, Bielefeld University, Germany
Volker Wendisch, Bielefeld University, Germany

Key Words: *Corynebacterium glutamicum*, volatile terpenoids, carotenoids, torulene.

Corynebacterium glutamicum is a natural carotenoid producing bacterium used in the million-ton-scale amino acid biotechnology that has been engineered for isoprenoid production[1]. The native membrane-bound carotenoid decaprenoxanthin is a rare C50 carotenoid. Volatile terpenoids such as valencene[2] and patchoulol[3] could be produced upon deletion of the first step of the specific carotenoid pathway and heterologous expression of the FPP synthase gene *ispA* from *E. coli* and terpene synthases from plant origin. However, these strains produced a yet unidentified carotenoid and only when all carotenoid biosynthetic genes were deleted, a colorless strain resulted. Expressing a codon optimized ADS from *Artemisia annua* in the white strain, amorphadiene, the volatile precursor for artemisinin was produced. For production of volatile terpenoids a dodecane overlay was used, a condition in which *C. glutamicum* benefits from its robust myco-membrane. Recently, we showed production of membrane-bound carotenoids with different length and/or cyclization status: bicyclic C50 sarcinaxanthin[4], bicyclic C40 astaxanthin[5], the linear lycopene[6] and the linear C50 bisanhydrobacterioruberin[7]. This indicated that the *C. glutamicum* myco-membrane accepts these linear and bicyclic carotenoids.
Here, we tested if the mono-cyclic C40 torulene can be produced by *C. glutamicum*. For this, a lycopene-overproducing strain was used as a platform strain to heterologously express a codon optimized lycopene cyclase/phytoene synthase gene *crtYB* from the torulene producing yeast *Sporidiobolus pararoseus*. This strain was analyzed in regard to its ability to transform lycopene into torulene under different membrane triggering conditions in order to enhance productivity of membrane-bound compounds.

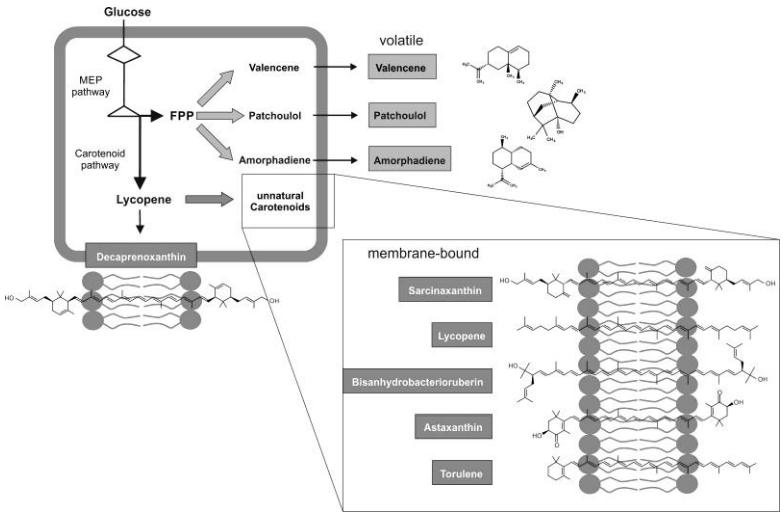

Figure 1 – Production of volatile and membrane-bound terpenoids with C. glutamicum.

[1] Heider SA, Wendisch VF. (2015) *Biotechnol J.* 10(8):1170-84.
[2] Frohwitter J, Heider SA, Peters-Wendisch P, Beekwilder J, Wendisch VF. (2014) *J Biotechnol.* 191:205-13.
[3] Henke NA, Wichmann J, Baier T, Frohwitter J, Lauersen KJ, Risse JM, Peters-Wendisch P, Kruse O, Wendisch VF. (2018) *Genes*. 9(4).
[4] Heider SA, Peters-Wendisch P, Netzer R, Stafnes M, Brautaset T, Wendisch VF. (2014) *Appl Microbiol Biotechnol.* 98(3):1223-35.
[5] Henke NA, Heider SA, Peters-Wendisch P, Wendisch VF. (2016) *Mar Drugs*. 14(7).
[6] Heider SA, Peters-Wendisch P, Wendisch VF. (2012) *BMC Microbiol.* 12:198.
[7] Taniguchi H, Henke NA, Heider SAE, Wendisch VF. (2017) *Metab Eng Commun.* 4:1-11.