Combinatorial Biosynthesis of Natural Products

THe LEGO-Principle to construct artifical biosynthetic pathways

Remco Muntendam, Verena Schütz, Arnim Quentmeier and Oliver Kayser

SCIENTIFIC HIGHLIGHTS

Plants and microorganisms are important sources for gene and drug discovery. Most isolated natural products with drug potential are structurally too complex, their isolation is too expensive or they can only be obtained in very low quantities. To overcome these problems, for combinatorial biosynthesis plant pathways are copied in microbial organisms being capable as biological factories to produce natural compounds of interest.

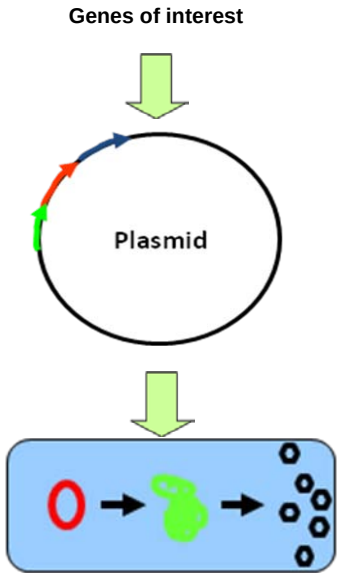
To engineer biosynthetic pathways we focus on terpenoids (*artemisinin*) and phenolics (*lignans*, *cannabinoids*) to produce them in microorganisms.

Terpene Factory. *Xanthophyllomyces dendrorhous* is a yeast well known for the industrial production of the orange food dye asthaxanthin. By genetic modification knock out lines are constructed to accumulate biosynthetic terpenoid precursors. By construction of genetic strains with multiple cloning cassettes we build universal strains for cloning genes of interest to biosynthesize non functionalized terpenoids.

Cannabinoid-Factory. Isolating or synthesizing tetrahydro-cannabinol (THC) as antiemetic drug for cancer patients is legally problematic or too expensive by organic synthesis. Biotechnological production in genetically modified baker yeast is a solution to this problem. We assemble the plant pathway, express heterologous genes and analyze host metabolom for optimal production yield. In addition we clone human liver cytochromes into plant and microbiological hosts to produce human metabolites for forensic chemistry.

***Cannabis sativa*, cultivated in Buitenpost, NL**

Lignan-Factory. Etoposide and tenoposide are two major anticancer drugs in clinics today. Both are semi-synthesized based on podophyllotoxin (PTOX) what is isolated from nearly extincted plants from the Himalaya region. *Anthriscus sylvestris* is an alternative common weed in Northern Europe producing lignans. By cloning a human cytochrome P450 in *A. sylvestris*, we modified the plant to produce PTOX. Plant secondary lignan metabolism is now under investigation and breeding studies are carried out with partners.
Additional isolation of endophytes showed a new approach for PTOX production. Endophytes are in plants symbiontic living fungi or bacteria producing secondary natural products like lignans. They are of main interest to understand the genetic blueprint and to learn about gene clustering.



Principle of combinatorial biosynthesis, cloning the genes of interest to suitable plasmid as vector, expression of genes in host and multiple enzymatic biocatalysis as biosynthesis of interest

oliver.kayser@bci.tu-dortmund.de
www.tb.bci.tu-dortmund.de

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