

Towards the metabolic engineering of myrcene pathway of *Pseudomonas sp. M1* using an integrated omic approach

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Pseudomonas sp. M1, isolated from the Rhine River, is able to utilize a large variety of toxic and/or recalcitrant compounds as sole carbon and energy sources, including phenols, benzene and monoterpenes like myrcene [1-3]. Therefore, M1 strain holds great potential as a source of novel biomolecules and cell factories for various biotechnological applications namely in biocatalysis, biosensors, bioremediation and biomedicine. However, the full exploitation of its enzymatic repertoire requires detailed and integrated information about the biomolecular catalog of M1 strain, including genes, proteins and metabolites.

In this context, the genome of *Pseudomonas sp. M1* was sequenced by *NGS technologies*, using Illumina Genome Analyser IIX and Roche 454 FLX. The resulting raw data was assembled into 41 contigs and annotated using different pipelines. The current genome draft of *Pseudomonas sp. M1* has an estimated GC content of 67%, a size of about 6.9 Mbps and includes 6214 CDS. Importantly, *in silico* genome analysis predicted a number of metabolic pathways involved in utilization/biotransformation of several unusual carbons sources (e.g. biphenyls, halophenols and different monoterpenes).

Proteomic and transcriptomic approaches have been setup envisaging the elucidation of the myrcene stimulon. In 2009, a set of myrcene-dependent proteins has been described using subproteome analysis of the cytoplasmic fraction [3]. More recently, a RNA-seq transcriptome analysis led to the identification of a 28kb genomic island of key importance in the catabolism of myrcene. This island includes genes involved in: i) myrcene oxidation and bioconversion of myrcene derivatives via a beta-oxidation like pathway; ii) regulation of myrcene pathway; iii) myrcene sensing. In addition several other gene clusters spread in the genome of *Pseudomonas sp. M1* have been found to be myrcene-dependently expressed and are currently being characterized.

Integration of genomic, transcriptomic, proteomic and metabolic data (which is currently being setup) will deliver a very solid and detailed description of the myrcene catabolism (and other monoterpenes), and on the associated molecular mechanisms of adaptation, providing the adequate support for the application of M1 as a biocatalyst in whole-cell biotransformations of plant-derived volatiles.

[1] Santos P.M., Sá-Correia I. (2007) *J. Biotechnol* **131**:371-378.

[2] Santos P.M., et al (2007) *OMICS: A Journal of Integrative Biology* **11**:233-251.

[3] Santos P.M., Sá-Correia I. (2009) *Proteomics* **9**:5101-11.

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