

## Abstract

Terpenoids are the largest class of natural products with remarkable chemical and structural diversity, making them a significant source of compounds for drug discovery. Terpenoids are used as food flavours, therapeutics, cosmetics, hormones or fuel. The starting scaffolds for terpenoid biosynthesis are condensed and, in some cases, cyclic carbohydrates synthesized from linear isoprenyl pyrophosphate precursors by terpene synthases. The enzyme family of terpene synthases is extensive and possesses well-characterized motifs and function-determining domains that can be used to search for homologous proteins with unknown catalytic function that might produce new compounds. Thanks to a bioinformatic pipeline developed in the Pluskal Lab in the Institute of Organic Chemistry and Biochemistry CAS, candidate terpene synthases with the potential to produce new terpene scaffolds were mined *in silico* from large protein sequence repositories. I expressed nine selected proteins originating from plants, bacteria, and fungi in the engineered budding yeast strains JWY501 and ZX178-08 that overproduce isoprenyl pyrophosphates. I then analysed the resulting yeast cell culture extracts using gas chromatography- and liquid chromatography-mass spectrometry. Five out of nine proteins exhibited terpene synthase activity and produced either di- or sesquiterpenes. Two sesquiterpenes produced by a fungal enzyme (UniProt id M2QMG2) were found to be potentially novel compounds, although further experiments are required to elucidate their molecular structures.