

Biosynthesis of Triacylglycerols in *Rhodococcus spp.*: Competition Between Metabolic Pathways of Different Storage Compounds

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Rhodococcus bacteria, which belong to the non-sporulating and mycolic acid-containing actinomycetes, are able to accumulate variable amounts of triacylglycerols (TAG) as main storage lipids. The application of microbial oils in the industry is currently a promising avenue; however, further basic and applied knowledge is required for making this biotechnological process economically competitive. To improve our general understanding of the relationship between TAG biosynthesis and other metabolic routes, we used different *Rhodococcus* strains, mutants defective in TAG biosynthesis and experiments with cerulenin, which is an inhibitor of the fatty acid biosynthesis. All studied strains, belonging to *R.opacus*, *R.jostii*, *R.erythropolis* and *R.fascians*, accumulated TAG as the main storage compounds, in addition to polyhydroxyalkanoates (PHA) (with 3-hydroxybutyrate and 3-hydroxyvalerate monomers) and glycogen (Gly) as minor compounds. A mutant of *R.opacus* PD630 with a knockout in one of the 10 *atf* genes (*atf1*), which codes for an acyltransferase responsible for TAG biosynthesis, showed significant decrease in TAG accumulation (approximately 50%) and an increase of approximately twice in the PHA and Gly contents. Other mutant of strain PD630 produced by chemical mutagenesis contained approximately four times increasing amounts of PHA and Gly, respectively, whereas the TAG accumulation decreased approximately seven times, compared with the wild type. When *R.opacus* was cultivated in the presence of cerulenin; the biosynthesis of TAG decreased drastically, whereas the cellular content of PHA and Gly increased twice and three times, respectively.

The results obtained in this study indicated that the biosynthesis routes of TAG, PHA and Gly in cells of *Rhodococcus spp.* compete for the carbon flux during cultivation of cells under nitrogen-limiting conditions. Whether the TAG accumulation by *Rhodococcus* members can be improved by metabolic engineering and re-direction of the carbon flux through metabolism, must be investigated in the future.