Identification of novel gene asset of *Rhodococcus opacus* R7 for polyethylene degradation by RNA-seq approach

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In the last two centuries, plastic played a revolutionary role for its versatile features, replacing other natural materials for packaging, transportation, storage, and garbage. The most used synthetic polymer is polyethylene (PE) for its chemical-physical and mechanical properties. Although the PE properties are favorable for its long lifetime, the same characteristic can lead to long-term impacts especially connected to PE waste disposal that still need to be addressed.

PE has always been considered as a chemically and biologically inert polymer, with a degradation yield (mechanical or biodegradation) that can occur only to a certain extent. PE mineralization is a complex process comprising the first oxidation of the hydrocarbon chain generating shorter aliphatic fragments that are consecutively degraded. Gram-negative and Gram-positives bacteria have been reported for the ability to degrade diverse types of PE, including few bacteria belonging to the *Rhodococcus* genus. Members of this genus are suitable for the biodegradation of recalcitrant contaminants since they possess unique adaptation capacities to fluctuating environmental conditions and to thrive under stress conditions. Consistently, they usually possess large genomes with redundant and versatile catabolic pathways.

In this scenario, an *omic*-approach contributed to unravel the complex degradative system behind PE biodegradation to better elucidate the gene system of *Rhodococcus opacus* R7. The strain is able to grow on PE used as the only carbon and energy source in a short range of time, increasing the total number of cells of almost two orders of magnitude. RNA-seq data analysis allowed to recover the genes involved in the first step of oxidation enabling the subsequent oxidation of short aliphatic chains. Specifically, genes putatively encoding for both oxidases and transport proteins were retrieved. A preliminary enzymatic assay provided insight into the gene category exerting the PE oxidation. Therefore, a laccase-like multi-copper oxidase (LMCO) was detected in R7 strain.

The identification of this marker sequence pose the basis for diverse applications: the biomonitoring of the quality of polluted environments with a culture-independent approach as well as biotechnological approach exploiting *R. opacus* R7 or the LMCO to biodegrade or biotransform PE wastes.