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Ether-lipid membrane engineering of *Escherichia coli*

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The membrane lipid composition of Archaea differs from Bacteria and Eukarya in having an ether-linked, isoprenoid hydrocarbon chain with enantiomer *sn*-glycerol-1-phosphate backbone. This unique structure is believed to be vital for the adaptation of these organisms to extreme environments and it reflects an evolutionary marker that distinguishes archaea from bacteria and eukaryotes. The aim of this project is to functionally introduce the archaeal ether-lipid biosynthetic pathway into *Escherichia coli* to create an Archeabacterium with improved tolerance to toxic compounds, to study the archaeal lipid biosynthesis and the evolutionary aspects associated with the lipid divide. Using a synthetic biology approach, upstream and downstream modules of the pathway were designed that consist of enzymes involved in the flux of the carbon source for the synthesis of the isoprenoid hydrocarbon chain and to synthesize CDP archaeol, the precursor for polar head group attachment. The latter ether-lipid is produced by a recent discovered gene encoding for CDP-archaeol synthase. The bacterial enzymes involved in the attachment of the L-serine and glycerol as polar head groups were examined due to their promiscuity in recognizing either archaeal or bacterial substrates. *In vivo* and *in vitro* assays were performed to access the enzymatic activities using chromatography and mass-spectrometry techniques in order to identify the archaeal ether lipids archaeatidylethanolamine (AE) and archaetidylglycerlol (AG). Using a combination of mutated bacterial and archaeal enzymes, these two archaeal lipids were produced in *E. coli*. Moreover, their formation can be reconstituted Ifrom basic building blocks and a set of purified enzymes.

Biography

Antonella Caforio has completed her Bachelor and Master studies at the University of Parma (Italy) and she is now at the final stage of her PhD in Molecular Microbiology at the University of Groningen (The Netherlands). Till now she has published three papers in scientific journals and one is still under review.

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