Search of mechanisms of adaptation to UV radiation in microorganisms from salterns using a functional metagenomics approach

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Abstract

One of the most damaging component to the microorganism is the solar UV radiation, which can be classified in three categories depend on the wavelengths: UV-A (315-400 nm), UV-B (280-315 nm) and UV-C (100-280 nm). The ozone absorbs most of the UV-C radiation which is the most harmful. Therefore, only a small amount of radiation can reach the Earth's surface. The microorganisms have developed molecular mechanisms to survive to this conditions. Nevertheless, the knowledge we have about those mechanisms are based on cultivable microorganisms. Thus, in order to find new mechanisms of adaptation to UV radiation damage, we use a functional metagenomics approach, which is an independent culture techniques. First, two metagenomic libraries were constructed from two samples of microorganisms of a solar saltern in Santa Pola (Alicante, Spain), named CR30 and CCAB, with 30% and 39% of salinity, respectively. Total DNA extracted from those microorganisms was partially digested with the Sau3A1 enzyme, and short DNA fragments (1-8 kb) were cloned into the plasmid pBluescript-SKII, using E. coli as host. The libraries were exposed on solid medium to UV-B radiation, under conditions that are lethal for the host E. coli strain. In summary, 8 fragments of environmental DNA (5 from CR30 and 3 from CCAB) were involved in UV resistance, a total of 20 genes from archaea and bacteria have been identified, of which 7 are responsible of the resistance phenotype. Those genes encodes for a 30S ribosomal protein S10, dihydrofolate reductase, response regulator, PAS sensor histidine kinase and the rest codify for hypothetical protein. Five of them were assigned to Haloquadratum walsbyi and two to Salinibacter ruber. Also, to test if the mechanism of UV resistance was related to protection against DNA damage, we used the chemical compound 4-nitroquinoline 1-oxide (4-NQO), which is considered a chemical analogue of UV radiation because it produces the same type of DNA lesions. Seven genes are resistant to both, UV and 4-NQO, and more genes are being tested now. We also have shown that the overexpression of the gene encoding for the ribosomal protein S10 in *Escherichia coli* gives resistance to UV an NQ. Finally, six of the genes also conferred resistance to perchlorate.

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