

52. Testing *Synechocystis* thylakoids as stores for recombinant membrane proteins

Mariano Battistuzzi¹, Mattia Niero¹, Anna Segalla¹, Francesca Zito², Bruno Miroux² and Elisabetta Bergantino¹

¹ *Department of Biology, University of Padova, Viale G.Colombo 3, 35121 Padova, Italy*

² *Institut de Biologie Physico-Chimique, UMR 7099, CNRS/Univerité Paris Diderot, Rue Pierre et Marie Curie 13, 75005 Paris, France*

Membrane proteins (MPs) characterization at the 3D level is by far lagging behind that of soluble proteins, essentially because of the difficulties in purifying them from native cells or tissue and also, alternatively, in over-producing them as recombinant products. A recent survey (Hattab et al., 2014) has shown that the T7 RNA polymerase-based expression system and the *E.coli* BL21(DE3) derivative strains C41 and C43, characterized by the proliferation of intracellular membranes upon overexpression of MPs (Miroux and Walker, 1996; Arechaga et al., 2000), account for more than 60 % of MP structures obtained after heterologous production. In these strains but also in other hosts, overproduction of lipids and development of a large network of internal membranes have proven to be extremely useful and productive in buffering the toxicity of MP recombinant expression.

We have recently improved a *Synechocystis* strain that represents the cyanobacterial counterpart of the *E.coli* strain BL21-C43, being similarly based on the induction of the T7 RNA polymerase and naturally possessing a wide internal membrane system, the thylakoids. We have planned to overexpress in it a palette of MPs including the *E.coli* ATPase b subunit to test its impact on membrane proliferation (as found by Miroux and Walker in 1996), bacteriorhodopsin (BR) as model protein and a mammalian mitochondrial uncoupling protein (UCP).

Arechaga & al (2000), FEBS Lett 482, 215-219.

Hattab & al (2014), in: Membrane Proteins Production for Structural Analysis. Mus-Veteau (Ed.), Springer-USA. Chapter 4, 87-106.

Miroux & Walker (1996), J Mol Biol 260, 289-298.