# Cisperone-Mediated Protein Folding *in vivo*: A New Platform Technology for High-Throughput Protein Expression

성백린*+, 신항철**, 최성일*
*Department of Biotechnology, Yonsei University and
**Department of Bioinformatics, Soongsil University, Seoul, KOREA
(Tel: 02 2123 2885; Fax: 02 362 7265)

A routine supply of functional proteins emerges as a new bottleneck for various disciplines of post-genome research initiatives - high-throughput protein crystallization for structure determination and functional analysis, development of new drug screening methods in combination with *in silico*design, and development of various therapeutic proteins (1, 2). A vexing problem for expression of recombinant protein in *E. coli* host is the inclusion body formation, precipitate of insoluble and inactive form of protein. Here, we report a new avenue for expression of proteins as soluble and functional forms by expeditingprotein folding in vivo.

The ProFuse vector system capitalizes on a new chaperone molecule, which facilitates foldingof target protein, when expressed as fusion to the chaperone. The chaperone functioning *in cis*,so christened with 'cisperone', is expected to work in a unique mechanism, distinct from the classical chaperones that work *in trans*. A variety of proteins were tested and the ProFuse vector displayed extreme ability for increasing the solubility of the expressed proteins, far exceeding conventional fusion vectors based on fusion to thioredoxin or MBP (3). The protein could be used for new assay development forenzyme inhibitor screening. Alternatively, after cleavage with site-specific proteases, the target protein could be purified as native form, suitable for HT crystallization studies and therapeutic applications.

References:

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2) Larsson, M. *et al. J. Biotechnol.* 80, 143 (2000)
3) Maina, C. V. *et al. Gene* 74, 365 (1988)