

Key stress factors and parameters for production optimisation of silk-elastin-like proteins in *E. coli* BL21(DE3)

Tony Collins, João Azevedo-Silva, André da Costa, Fernando Branca, Raul Machado, Margarida Casal

tcollins@bio.uminho.pt

Centre of Molecular and Environmental Biology (CBMA), Department of Biology, University of Minho, Braga, Portugal

Abstract

Silk-elastin-like proteins (SELPs) combining the physicochemical and biological properties of silk and elastin have a high potential for use in the pharmaceutical, regenerative medicine and materials fields. Their development for use is however restrained by their production levels. We have recently synthesised a series of novel silk-elastin-like proteins and here we will describe the optimisation of the production of these with the pET-*E. coli* BL21(DE3) expression system. Both batch production in shake flasks and fed-batch production approaches were investigated. Furthermore, a comprehensive empirical approach examining all process variables (media, medium composition, inducer, induction time and period, temperature, pH, aeration, agitation, pre- and post-induction growth rates) and a detailed characterisation of the bioprocesses were carried out in an attempt to maximise production and to identify the factors limiting higher production levels. Using the optimised conditions, approximately 0.5 g/l of purified SELP was obtained in shake flasks and as much as 4 g/L was obtained when using the fed-batch approach. These represent, respectively, approximately 10 and almost 100-fold increases on that previously reported for SELPs.

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Keywords: protein based polymers, silk-elastin-like polymers, production optimisation, batch and fed-batch production, pET-*E. coli* BL21(DE3).

References

Collins T., Azevedo-Silva J., da Costa A., Branca F., Machado R. and Casal M. (2013). Batch production of a silk-elastin-like protein in *E. coli* BL21(DE3): key stress factors and parameters for optimisation. *Microb. Cell Fact.* Feb 27; 12:21. doi: 10.1186/1475-2859-12-21.

Machado M., Correia C., Azevedo-Silva J, Collins T, Arias J, Rodríguez-Cabello J.C. and Casal M. (2013). High level expression and facile purification of recombinant silk-elastin-like polymers in auto induction shake flask cultures. *AMB Express.* Feb 5; 3(1):11. doi: 10.1186/2191-0855-3-11.