Cent. Eur. J. Biol. • 8(1) • 2013 • 1-7
DOI: 10.2478/s11535-012-0112-6

VERSITA

Central European Journal of **Biology**

Comparison of signal peptides for efficient protein secretion in the baculovirus-silkworm system

Research Article

Yasuhiko Soejima[1], Jae Man Lee[1], Yudai Nagata[1], Hiroaki Mon[1], Kazuhiro Iiyama[2], Hajime Kitano[3], Michiya Matsuyama[3], Takahiro Kusakabe[1,*]

[1]*Laboratory of Silkworm Sciences, Faculty of Agriculture, Kyushu University, 812-8581 Fukuoka, Japan*

[2]*Laboratory of Insect Pathology, Faculty of Agriculture, Kyushu University, 812-8581 Fukuoka, Japan*

[3]*Laboratory of Marine Biology, Faculty of Agriculture, Kyushu University, 812-8581 Fukuoka, Japan*

Received 21 March 2012; Accepted 26 September 2012

Abstract: The baculovirus-silkworm expression system is widely used as a mass production system for recombinant secretory proteins. However, the final yields of some recombinant proteins are not sufficient for industrial use. In this study, we focused on the signal peptide as a key factor for improving the efficiency of protein production. Endoplasmic reticulum (ER) translocation of newly synthesized proteins is the first stage of the secretion pathway; therefore, the selection of an efficient signal peptide would lead to the efficient secretion of recombinant proteins. The *Drosophila* Bip and honeybee melittin signal peptides have often been used in this system, but to the best of our knowledge, there has been no study comparing secretion efficiency between exogenous and endogenous signal peptides. In this study we employed signal peptides from 30K Da and SP2 proteins as endogenous signals, and compared secretion efficiency with those of exogenous or synthetic origins. We have found that the endogenous secretory signal from the 30K Da protein is the most efficient for recombinant secretory protein production in the baculovirus-silkworm expression system.

Keywords: *Baculovirus expression system • Silkworm • Signal peptide • Secretion • Protein purification*

1. Introduction

The baculovirus-silkworm expression system has several advantages over other recombinant protein expression systems in producing recombinant secretory proteins [1]. Most importantly, this system permits, at least in part, posttranslational modifications of proteins found in mammalian cells [2,3]. Under the control of a strong polyhedrin promoter, recombinant proteins are expressed at extremely high levels and accumulate within a cell or are secreted into hemolymph [1]. However, in some cases, a protein of interest is secreted at a low level and a large amount of the protein remains in the cell. One possible explanation for this observation is that some or all of the secreted proteins were unable to translocate into the endoplasmic reticulum (ER) lumen. The N-terminal signal peptide is a key factor in determining the efficiency of ER translocation from ribosome to ER membrane. Several studies have demonstrated that recombinant proteins can be produced with a high yield by replacing signal peptides in lytic and nonlytic insect expression systems [4-6]. However, there have been few reports regarding the optimal signal peptide that facilitates efficient protein secretion in the baculovirus-silkworm system, which is a high-scale expression system compared with the insect cell system.

A signal peptide consists of three regions: a positively charged amino-terminal region (N-region, 1-5 residues), a central hydrophobic region (H-region, 7-15 residues), and a more polar carboxy terminal region (C-region, 3-5 residues) [7,8]. Although the compositions of amino acids and lengths of signal peptides are highly diverse among protein species, the overall structure of these three functional regions is conserved [9]. During nascent peptide-chain synthesis on ribosomes bound to the ER membrane, a signal recognition particle (SRP) binds

* *E-mail: kusakabe@agr.kyushu-u.ac.jp*