Technology Offer

Fast and efficient protein purification form prokaryotic and eukaryotic hosts

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Fusion Tags Comprising Ubiquitin-like Modifiers and Corresponding Proteases For Protein Purification From Selected Eukaryotic Expression Hosts

Background

Affinity tags allow for a fast and streamlined purification of recombinant proteins. Ideally, however, such tags are proteolytically removed during the purification procedure. Recently, particular advantageous affinity tag modules comprising SUMO or other eukaryotic ubiquitin-like modifiers (UBLs) such as NEDD8, Atg8, or ubiquitin as specific protease recognition sites have been introduced (1, 2). Such UBL-containing fusion tags often facilitate protein expression and solubility, and can be efficiently cleaved in a site-specific manner by corresponding UBL-specific proteases. However, despite the availability of various UBL-containing fusion tags with different protease cleavage sites, their application has mostly been restricted to prokaryotic expression hosts, because in eukaryotic expression hosts they are typically rapidly cleaved *in vivo* by endogenous UBL-specific proteases. Currently, only one engineered variant of SUMO (SUMOstar from LifeSensors) and a correspondingly optimized protease (SUMOstar Protease from LifeSensors) are applicable to purification strategies using eukaryotic expression hosts.

Technology

Scientists from the Max-Planck Institute for Biophysical Chemistry in Göttingen have recently expanded the list of UBL-containing fusion tags and corresponding UBL-specific proteases applicable to purification strategies for proteins and protein complexes expressed in both prokaryotic and selected eukaryotic hosts.
The researchers demonstrated that affinity tags comprising xlLC3B from *X. laevis* or bdNEDD8 from *B. distachyon* are resistant to *in vivo* cleavage not only in prokaryotes (*E. coli*) but also in selected eukaryotic expression hosts. For example, xlLC3B or bdNEDD8 fusion proteins can be produced in fungal hosts like *S. cerevisiae*. In addition, xlLC3B fusions are stable in wheat germ extract as well as in the presence of recombinant wheat Atg4. xlLC3B is therefore a promising partner for production of recombinant fusion proteins not only in fungi but also in plants. Both protease recognition sites (xlLC3B and bdNEDD8) can be efficiently cleaved in downstream purification procedures by the corresponding xlLC3B-specific protease xlAtg4 or the bdNEDD8-specific protease bdNEDP1, respectively. Both xlAtg4 and bdNEDP1 are applicable to both, on-column and post-column digests, have a particular high efficacy over a broad temperature range (0°C-37°C), and are characterized by a high salt-tolerance (up to at least 1 M NaCl).

We are currently seeking for a partner who is interested in licensing and commercializing these new affinity tag modules and/or the corresponding proteases.

Max-Planck-Innovation GmbH
Amalienstr. 33
80799 München
www.max-planck-innovation.de

Patent Information

A European priority application has been filed in November 2014

Literature

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(2) Frey S and Goerlich D, *Journal of Chromatography A*, 1337 (2014), 95-105

Contact

Dr. Dieter Link

Senior Patent- & License Manager
Biologist

Phone: +49 (0)89 / 29 09 19 - 28
eMail: dieter.link@max-planck-innovation.de

Max-Planck-Innovation GmbH
Amalienstr. 33
80799 München
www.max-planck-innovation.de