

论文

串联亲和纯化技术筛选hCLP46的相互作用蛋白

徐峰¹, 穆昕², 王崑¹, 刘利新¹

1. 中国科学院研究生院, 北京100049;
2. 中国科学院生物物理研究所, 北京 100101

摘要:

hCLP46(human CAP10-like protein46)是从MDS-AML患者的CD34⁺干细胞cDNA文库中筛选出的基因.我们利用串联亲和纯化技术来筛选与hCLP46有相互作用的蛋白.通过体内交联-甘氨酸洗脱策略,检测到7条有差异的蛋白带.经液相色谱-质谱联用鉴定,得到了CNX和PDI等一系列内质网伴侣蛋白.所以hCLP46可能是一个糖蛋白,其成熟过程利用了BiP/Grp94和CNX/CRT 2套伴侣蛋白系统.

关键词: 交联剂 串联亲和层析 骨髓增生异常综合症 hCLP46蛋白

Screening proteins interacting with hCLP46 using tandem affinity purification

XU Feng¹, MU Xin², WANG Wei¹, LIU Li-Xin¹

1. Graduate University, Chinese Academy of Sciences, Beijing 100049, China;
2. Institute of Biophysics, Chinese Academy of Sciences, Beijing 100101, China

Abstract:

hCLP46 (human CAP10-like protein46), a novel gene, has been screened out from the cDNA library of MDS-AML Patient's CD34⁺ stem cell. For to further studying the biological characteristics of this gene tandem affinity purification (TAP) has been used to isolate proteins which specifically interact with hCLP46, and seven specific protein bands were detected. By using liquid chromatography-mass spectrometry and protein database searching, a series of endoplasmic reticulum chaperones were identified. Two major ER chaperone systems, the BiP/Grp94 and the calnexin (CNX)/ calreticulin (CRT) systems, are important in the maturation of hCLP46.

Keywords: crosslinker tandem affinity purification(TAP) MDS hCLP46

收稿日期 2010-05-24 修回日期 2010-05-27 网络版发布日期

DOI:

基金项目:

国家自然科学基金(30670889,30771193)资助

通讯作者:

作者简介:

作者Email: lxliu@gucas.ac.cn

参考文献:

- [1] Teng Y, Liu Q, Ma J, et al. Cloning, expression and characterization of a novel human CAP10-like gene *hCLP46* from CD34⁺ stem/progenitor cells [J]. *Gene*, 2006, 371: 7-15.
- [2] Ponting C P, Mott R, Bork P, et al. Novel protein domains and repeats in *Drosophila melanogaster*: Insights into structure, function, and evolution [J]. *Genome Research*, 2001, 11: 1996-2008.
- [3] Acar M, Jafar-Nejad H, Takeuchi H, et al. Rumi is a CAP10 domain glycosyltransferase that modifies notch and is required for notch signaling

扩展功能

本文信息

- ▶ Supporting info
- ▶ PDF(1659KB)
- ▶ [HTML全文]
- ▶ 参考文献[PDF]
- ▶ 参考文献

服务与反馈

- ▶ 把本文推荐给朋友
- ▶ 加入我的书架
- ▶ 加入引用管理器
- ▶ 引用本文
- ▶ Email Alert
- ▶ 文章反馈
- ▶ 浏览反馈信息

本文关键词相关文章

- ▶ 交联剂
- ▶ 串联亲和层析
- ▶ 骨髓增生异常综合症
- ▶ hCLP46蛋白

本文作者相关文章

PubMed

[J]. Cell, 2008, 132: 247-258.

[4] Zanotti S, Canalis E. Notch and the Skeleton

[J]. Molecular and Cellular Biology, 2010, 30: 886-896.

[5] Rigaut G, Shevchenko A, Rutz B, et al. A generic protein purification method for protein complex characterization and proteome exploration

[J]. Nature Biotechnology, 1999, 17: 1030-1032.

[6] Goto H, Kawaoka Y. A novel mechanism for the acquisition of virulence by a human influenza A virus

[J]. Proceedings of the National Academy of Sciences of the United States of America, 1998, 95:

10224-10228.

[7] Haas I G, Wabl M. Immunoglobulin heavy chain binding protein

[J]. Nature, 1983, 306: 387-389.

[8] Lievremont J P, Rizzuto R, Hendershot L, et al. BiP, a major chaperone protein of the endoplasmic reticulum lumen, plays a direct and important role in the storage of the rapidly exchanging pool of Ca^{2+}

[J]. J Biol Chem, 1997, 272: 30873-30879.

[9] Kabani M, Kelley S S, Morrow M W, et al. Dependence of endoplasmic reticulum-associated degradation on the peptide binding domain and concentration of BiP

[J]. Mol Biol Cell, 2003, 14: 3437-3448.

[10] Molinari M, Helenius A. Chaperone selection during glycoprotein translocation into the endoplasmic reticulum

[J]. Science, 2000, 288: 331-333.

[11] Ellgaard L, Helenius A. ER quality control: towards an understanding at the molecular level

[J]. Current Opinion in Cell Biology, 2001, 13: 431-437.

[12] Hebert D N, Molinari M. In and out of the ER: Protein folding, quality control, degradation, and related human diseases

[J]. Physiological Reviews, 2007, 87: 1377-1408.

[13] Helenius A, Aebi M. Roles of N-linked glycans in the endoplasmic reticulum

[J]. Annu Rev Biochem, 2004, 73: 1019-1049.

[14] Trombetta E S, Simons J F, Helenius A. Endoplasmic reticulum glucosidase II is composed of a catalytic subunit, conserved from yeast to mammals, and a tightly bound noncatalytic HDEL-containing subunit

[J]. Journal of Biological Chemistry, 1996, 271: 27509-27516.

[15] Chevet E, Jakob C A, Thomas D Y, et al. Calnexin family members as modulators of genetic diseases

[J]. Seminars in Cell and Developmental Biology, 1999, 10: 473-480.

[16] Cai H, Wang C C, Tsou C L. Chaperone-like activity of protein disulfide-isomerase in the refolding of a protein with no disulfide bonds

[J]. Journal of Biological Chemistry, 1994, 269: 24550-24552.

[17] Quan H, Fan G B, Wang C C. Independence of the chaperone activity of protein disulfide-isomerase from its thioredoxin-like active-site

[J]. Journal of Biological Chemistry, 1995, 270: 17078-17080.

[18] Maattanen P, Kozlov G, Gehring K, et al. ERp57 and PDI: multifunctional protein disulfide isomerases with similar domain architectures but differing substrate-partner associations

[J]. Biochemistry and Cell Biology-Biochimie Et Biologie Cellulaire, 2006, 84: 881-889.

[19] Oliver J D, Roderick H L, Llewellyn D H, et al. ERp57 functions as a subunit of specific complexes formed with the ER lectins calreticulin and calnexin

[J]. Molecular Biology of the Cell, 1999, 10: 2573-2582.

[20] Chevet E, Wong H N, Gerber D, et al. Phosphorylation by CK2 and MAPK enhances calnexin association with ribosomes

[J]. Embo Journal, 1999, 18: 3655-3666.

本刊中的类似文章

Copyright by 中国科学院研究生院学报