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[1] 邹菊, 刘志刚. 大豆主要过敏原Gly m Bd 30K蛋白单克隆抗体的制备与应用[J]. 大豆科学, 2011, 30(05): 723-726. [doi:10.11861/j.issn.1000-9841.2011.05.0723]
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大豆主要过敏原Gly m Bd 30K蛋白单克隆抗体的制备与应用

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摘要: 利用大豆主要过敏原Gly m Bd 30K蛋白抗原表位蛋白为免疫原免疫BALB/c小鼠, 取免疫小鼠脾细胞与小鼠骨髓瘤NS-1细胞融合。采用半固体培养基法和有限稀释法相结合的方法快速筛选获得稳定分泌的特异性杂交瘤细胞, 用杂交瘤细胞株产生小鼠腹水, 应用蛋白A亲和层析法进行抗体纯化。采用Ig类与亚类鉴定试剂盒鉴定该单克隆抗体的Ig亚型; 通过间接ELISA、Western Blotting鉴定该单克隆抗体的特性和交叉性。利用双单抗夹心ELISA法检测大豆过敏原。结果表明: 获得6株可稳定分泌鼠抗大豆主要过敏原Gly m Bd 30K蛋白的单克隆抗体, 分别命名为1C10, 1D12, 2D1, 4B4, 5F9, 6B12, 其Ig亚型除1D12和4B4为IgG2a外, 其余均为IgG1, 且6株单抗效价均在10⁵以上。ELISA和Western Blotting分析表明该6株单抗均能特异性识别大豆主要过敏原Gly m Bd 30K蛋白, 并且建立双单抗夹心ELISA的方法可以准确检测出大豆过敏原的存在。鼠抗大豆主要过敏原Gly m Bd 30K蛋白抗原表位区蛋白的单克隆抗体的成功制备, 以及双单抗夹心ELISA检测系统的建立, 为大豆主要过敏原蛋白的检测奠定了基础, 也可以为食品中大豆过敏原的检出提供依据。

Abstract: To prepare monoclonal antibodies against allergen soybean, BALB/c mice were immunized with antigenic epitope of Gly m Bd 30K protein from soybean, and the splenocytes of the immunized mice were fused with NS-1 myeloma cells by hybridoma technique. The McAbs were purified using affinity chromatography on immobilized protein A and identified by their specificity, subtype, titers and cross-reactivity with ELISA and Western blotting. With the method of sandwich-ELISA to detect soybean allergen protein trace in food products. Six hybridoma cell lines secreting McAbs against antigenic epitope of Gly m Bd 30K protein from soybean were obtained, which were denominated as 1C10, 1D12, 2D1, 4B4, 5F9, 6B12. The six McAbs all recognized recombinant antigenic epitope of Gly m Bd 30K protein from soybean. A sandwich-ELISA system was set up to detect the presence of soybean allergens. These six monoclonal antibodies against antigenic epitope of Gly m Bd 30K protein from soybean were prepared successfully, which would facilitate establishing detection method of soybean allergen.

参考文献/References:

- [1] Metcalfe D. The nature and mechanisms of food allergies and related diseases[J]. Food Technology, 1992, 5(5): 136-140.
- [2] Duke W W. Soybean as a possible important source of allergy[J]. Journal of Allergy, 1934, 5: 300-302.
- [3] 李钢, 丁淑霞, 左容, 等. 食物过敏引起变态反应疾病184例临床分析[J]. 佳木斯医学院学报, 1989, 12(3): 269. (Li G, Ding S X, Zuo R, et al. Food allergies cause allergic disease clinical analysis of 184 cases[J]. Jiamusi Medical College, 1989, 12(3): 269.)
- [4] Dean D M, Hugh A S, Ronald A S. Food allergy: adverse reactions to foods and food additives (Second Edition) [M]. Massachusetts: Blackwell Publishing, 1997: 253-267.
- [5] 周淑红. 国外关于食品过敏标签的现状与启示[J]. 世界农业, 2007(6): 67-68. (Zhou S H. The apocalypse and status on food allergy labels in abroad[J]. World Agriculture, 2007(6): 67-68.)
- [6] 陈家杰, 朱海, 叶卫翔, 等. 双抗体夹心ELISA法测定食物中大豆过敏原蛋白成分[J]. 食品研究与开发, 2009, 30(5): 105-109. (Chen J J, Zhu H, Ye W X, et al. Detection of soybean allergen protein trace in food products by sandwich-antibody enzyme linked immunosorbent assay[J]. Food Research and Development, 2009, 30(5): 105-109.)
- [7] Ogawa T, Bando N, Tsuji H, et al. Investigation of the IgE binding proteins in soybeans by immunoblotting with the sera of the soybean sensitive patients with atopic dermatitis[J]. Journal of Nutritional Science and Vitaminology, 1991, 37(6): 555-565.

- [8]Kalinski A J, Melroy D L, Dwivedi R S, et al. A soybean vacuolar protein(P34) related to thiol proteases which is synthesized as a glycoprotein precursor during seed maturation[J]. Journal of Biological Chemistry, 1992, 267(17):12068-12076.
- [9]Kalinski A J, Weisemann J, Matthews B F, et al. Molecular cloning of a protein associated with soybean oil bodies which is homologous to thiol proteases of the papain family[J]. Journal of Biological Chemistry, 1990, 265(23):13843-13848.
- [10]Ogawa T, Tsuji H, Kitamura K, et al. Identification of the soybean allergenic protein, Gly m Bd 30 K, with the soybean seed 34-kDa oil-body-associated protein[J]. Bioscience, Biotechnology and Biochemistry, 1993, 57(6):1030-1033.
- [11]邹玉兰, 刘志刚. 大豆主要过敏原Gly m Bd 30K基因的克隆及其原核表达载体的构建[J]. 大豆科学, 2009, 28(1):11-15. (Wu Y L, Liu Z G. Cloning and prokaryotic expression vector construction of Gly m Bd 30K gene from soybean(Glycine max) [J]. Soybean Science, 2009, 28(1):11-15.)
- [12]林苏霞, 王晓梅, 刘志刚, 等. 大豆主要过敏原Gly m Bd 30K的抗原表位区基因的克隆表达、纯化及免疫原性鉴定[J]. 大豆科学, 2010, 29(2):186-190. (Lin S X, Wang X M, Liu Z G, et al. Cloning and expression of the antigenic epitope of Gly m Bd 30K protein from soybean and purification and identification of expressed product [J]. Soybean Science, 2010, 29(2):186-190.)
- [13]游金明, 王自蕊, 谯仕彦, 等. 致仔猪过敏性大豆抗原蛋白 β conglycinin单克隆抗体的制备与鉴定[J]. 中国畜牧杂志, 2008, 44(17):46249. (You J M, Wang Z R, Qiao S Y, et al. Preparation and characterization of monoclonal antibodies against soybean β -conglycinin, a hypersensitive allergen to piglets [J]. Chinese Journal of Animal Science, 2008, 44(17):46-49.)
- [14]邹菊, 张强, 刘志刚. 抗鸡蛋卵类黏蛋白单克隆抗体的制备与鉴定[J]. 免疫学杂志, 2011, 27(3):185-188. (Zou J, Zhang Q, Liu Z G. Preparation and potential application of monoclonal antibodies against allergen ovomucoid [J]. Immunological Journal, 2011, 27(3):185-188.)
- [15]陈嵘祯, 廖家, 林映萍, 等. 结节性痒疹患者血清中食物不耐受sIgG、IgG亚型和TNF- α 水平测定[J]. 广东医学院学报, 2008, 26(6):597-598. (Chen R Y, Liao J, Lin Y P, et al. Serum levels of food intolerance-specific IgG, IgG subtypes and TNF- α in prurigo nodularis [J]. Journal of Guangdong Medical College, 2008, 26(6):597-598.)

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