

农产品辐照研究·食品科学

特布他林杂交瘤细胞株的建立及其单克隆抗体制备与鉴定

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摘要:

分别通过1,4-丁二酐法和EDC法将特布他林偶联于载体蛋白BSA和OVA上,用BSA-TBL免疫BALB/c小鼠,经过3次免疫后,OVA-TBL包被后用间接ELISA和阻断ELISA选择细胞融合备用鼠,选择高效价、高敏感性和高特异性的小鼠进行抗原进行冲击免疫;无菌手术取其脾细胞与骨髓瘤细胞融合建立分泌TBL 单克隆抗体的杂交瘤细胞株;采用体内诱生腹水法制备TBL mAb,并对TBL mAb的效价、敏感性和特异性等免疫学特性进行鉴定。结果显示,免疫的6只小鼠血清抗体效价均达到 $10^{-4}$ ;融合后筛选出4C08-G5和3H3-A02共2株敏感特异的杂交瘤细胞,其细胞培养上清液效价分别为1 : 800和1 : 1600,腹水效价分别为1 :  $2.56 \times 10^5$ 和1 :  $1.02 \times 10^6$ ; 4C08-G5株分泌的抗体对TBL的 $IC_{50}$ 为5.25ng/ml,与瘦肉精、莱克多巴胺等其他 $\beta_2$ 激动剂交叉反应性小于3%。本试验获得了抗TBL mAb,为TBL残留免疫检测方法的建立奠定了坚实的基础。

关键词: 特布他林 杂交瘤 单克隆抗体 鉴定

ESTABLISHMENT OF HYBRIDOMA CELL LINES SECRETING ANTI-TERBUTALINE MONOCLONAL ANTIBODY AND PREPARATION, IDENTIFICATION OF ITS IMMUNOLOGICAL PROPERTIES

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Abstract:

Artificial antigen BSA-TBL(terbutaline) and OVA-TBL were synthesized using butane-1,4-diol diglycidyl ether and EDC by linking carrier proteins BSA and OVA to TBL. The titers of polyclonal antibody were detected by indirect ELISA and blocking ELISA after three times immunization by BSA-TBL. The high titer, sensitivity and specificity mouse was selected for cell fusing. The hybridoma lines that secrete TBL mAb were established with using monoclonal antibody hybridoma technology and the immunological characteristics such as titer, sensitivity and specificity of the mAb were characterized. The results showed that six BALB/c mice indirect ELISA titer against TBL were above  $1 \times 10^{-4}$  and three hybridoma cell lines of 4C08-G5 and 3H3-A02 were screened for specificity to TBL, the indirect ELISA titer of the mAb were 1 : 800 and 1 : 1600 in supernatant, 1 :  $2.56 \times 10^5$  and 1 :  $1.02 \times 10^6$  in ascites, the mAb of 4C08-G5 showed good sensitivity with an  $IC_{50}$  of 5.25 ng/ml to TBL and had less than 30% cross-reactivity to other compounds. The results showed TBL mAb has been generated and made it possible to establish immunoassay of TBL residues.

Keywords: Terbutaline hybridoma monoclonal antibody characterization

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