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重组人红细胞生成素中N-连接糖的毛细管凝胶电泳-激光诱导荧光测定法

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摘要:

目的:表征重组人红细胞生成素中影响体内生物学活性的N-连接糖谱。方法:以毛细管凝胶电泳-激光诱导荧光检 测法(CGE-LIF)测定了重组人红细胞生成素(rHuEPO)中N-连接的多个糖基化形式,并以此为基础研究结构与活性 的关系。结果:同一生产线上的样品,其N-连接糖的CGE-LIF图谱基本一致;不同表达载体所表达的rHuEPO,其 N-连接糖的形式不同;活性不同的样品,各N-连接糖组分的相对含量存在很大差异。结论:建立的方法可用于判断 产品的来源及批间差异,与肽图相结合可测定rHuEPO的一级结构。

关键词: 毛细管凝胶电泳-激光诱导荧光检测法; 重组人红细胞生成素; N-连接糖

CHARACTERIZATION OF N-GLYCANS FROM RECOMBINANT HUMAN FRYTHROPOLETIN BY CAPILLARY ELECTROPHORESIS WITH LASER-INDUCED FLUORESCENCE

Zhou Guohua; Luo Guoanand Zhang Xiaodan

Abstract:

AIM: To characterize the profiles of N-glycans in highly glycosylated protein recombinant human erythropoietin (rHuEPO). METHODS: Capillary gel electrophoresis (CGE) of sugars in recombinant human pg国安 erythorpoietin derivatized by reductive amination with 8-aminopyrene-1,3,6-trisulfonate(APTS) and detected by laser-induced fluorescence (LIF) has been performed to study the relationships between structure and bioactivities of rHuEPO. RESULTS: The results showed that the CGE-LIF profiles of Nglycans in rHuEPO from the same cell-line were almost identical, but clear different profiles emerged when the sample was from different expression vector. Significant differences in CGE-LIF mappings of rHuEPO with different in vivo bioactivities were also observed. CONCLUSION: The approach developed in this article can be used to control the first order structure of rHuEPO by combining with peptide mapping.

Keywords: recombinant human erythropoietin N-glycans capillary gel electrophoresis with laserinduced fluorescence

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