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论文

抗体夹心酶联免疫吸附法测定重组*E.coli L*-天冬酰胺酶及药代动力学研究

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摘要:

目的建立大鼠血浆中重组*E.coli L*-天冬酰胺酶的抗体夹心酶联免疫吸附测定法,并进行药代动力学研究。方法应用重组*E.coli L*-天冬酰胺酶免疫家兔,分离IgG,用DEAE-纤维素柱色谱纯化,辣根过氧化物酶标记抗体,建立抗体夹心酶联免疫吸附法,测定大鼠血浆中重组*E.coli L*-天冬酰胺酶浓度。结果方法的线性范围为1～64 $U\cdot L^{-1}$,血药浓度与时间的关系符合二房室模型,初期和末端的$T_{1/2}$分别为0.50～0.57 h和2.45～3.02 h,AUC与剂量成正比。结论建立的抗体夹心酶联免疫吸附法在灵敏度、特异性、线性范围、精密度和回收率等方面,满足药代动力学研究要求。实验方法和重组*E.coli L*-天冬酰胺酶在大鼠中的药代动力学参数为临床研究提供了手段和依据。

关键词: 重组*E.coli L*-天冬酰胺酶 辣根过氧化物酶标记抗体 抗体夹心酶联免疫吸附法 药代动力学

Antibody sandwich enzyme-linked immunoadsorbent assay for determination of recombinant *E.coli L*-asparaginase in rat plasma and its pharmacokinetics

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Abstract:

AimTo establish antibody sandwich enzyme-linked immunoadsorbent assay for determination of recombinant *E.coli L*-asparaginase in rat plasma and study its pharmacokinetics. MethodsA Japanese white rabbit was immunized with recombinant *E.coli L*-asparaginase. Immunoglobulin G was separated and purified by using DEAE-cellulose chromatography. Conjugation of horseradish peroxidase to immunoglobulin G was obtained using the two-step glutaraldehyde method. Recombinant *E.coli L*-asparaginase protein in plasma was measured by antibody sandwich enzyme-linked immunoadsorbent assay. Pharmacokinetic parameters were assessed with model-dependent method. ResultsThe linearities was 1-64 $U\cdot L^{-1}$. Concentration-time profile after iv of 1.250, 2.50, 5.00 $kU\cdot kg^{-1}$ of recombinant *E.coli L*-asparaginase fitted with a two-compartment model. The first and terminal elimination $T_{1/2}$ were 0.50-0.57 h and 2.45-3.02 h, respectively. The AUC was linearly related to the doses. ConclusionAntibody sandwich enzyme-linked immunoadsorbent assay was constant, reliable, sensitive, and suitable for the determination of recombinant *L*-asparaginase. Pharmacokinetics of recombinant *E.coli L*-asparaginase in rats is warranted for the design of future clinical trails.

Keywords: IgG conjugated horseradish peroxidase antibody sandwich enzyme-linked immunoadsorbent assay pharmacokinetics recombinant *E.coli L*-asparaginase

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