

畜牧兽医科学

鸡IL-2的毕赤酵母表达和生物活性鉴定

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摘要: IL-2是一种重要的免疫相关细胞因子,借助PCR去掉鸡IL-2基因片段(387bp)信号肽,连接到pPICZaA表达载体中,通过电转化转入Pichia酵母GS115菌株的感受态中,对筛选出来的阳性菌株以终浓度为1%的甲醇诱导表达,经SDS-PAGE及Western Blot分析,结果表明酵母成功分泌表达了重组鸡IL-2,分子量为17.8kD。通过Ni-NTA His Bind 柱层析,250mM咪唑的洗脱液洗脱,得到纯度较高的重组鸡IL-2,淋巴细胞增殖实验证明,重组鸡IL-2具有明显的促进淋巴细胞增殖的生物活性,为进一步研究其免疫佐剂的作用提供了实验基础。

关键词: 鸡IL-2, pPICZaA表达载体, 毕赤酵母, 分泌表达

Expression in Pichia pastoris and biological activity characterization of recombinant chicken IL-2

Abstract: Chicken IL-2 is an key cytokine for immunity. Removed the signal peptide by PCR, chIL-2 gene fragments (387bp) was connected to the pPICZaA expression vector and transformed to Pichia pastoris GS115 by electroporation. SDS-PAGE and Western Blot analysis showed that chIL-2(17.8kD) was successfully expressed and secreted by positive strains induced by 1% methanol. The chIL-2, which was purified by Ni-NTA His Bind column chromatography with 250mM imidazole elution buffer, had significant biological activity to promote lymphocyte proliferation, which provides the experimental basis for the further research on chIL-2 as a kind of immune adjuvant.

Keywords: Chicken IL-2, pPICZaA expression vector, Pichia pastoris, Secretary expression

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