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基于生物素·亲和素放大的SPR传感器检测大肠杆菌研究

作 者: 刘儒平, 王程, 徐万帮, 岳钊, 牛文成, 刘国华

单 位: 南开大学信息技术科学学院

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摘 婁

建立了一种基于表面等离子体共振(SPR)技术的大肠杆菌特异性快速检测方法。采用EDC/NHS将葡聚糖修饰的CM5芯片表面活化,通过亲和素固定生物素标记的二抗(羊抗兔IgG抗体),利用一抗(兔抗大肠杆菌 ATCC25922单克隆抗体)和二抗反应,将兔抗大肠杆菌单克隆抗体固定在传感芯片表面。利用一抗和二抗的质量扩增效应和生物素-亲和素的多级放大效应,实现了对低浓度大肠杆菌 ATCC25922的快速检测,提高了SPR传感器灵敏度。利用NaOH溶液对芯片再生,可对多个不同浓度样品进行检测,采用相对响应单位(RU)记录数据。本传感芯片对大肠杆菌 ATCC25922响应的线性范围为1.5×10/2~1.5×10/7 CFU/mL,检测限为1.5×10/2 CFU/mL,相关系数r为0.9815。这种方法简便快捷,有望成为一种在线检测治病菌的有力手段。

关键词: SPR传感器; 生物素-亲和素多级放大; 大肠杆菌 ATCC25922; 快速检测

Streptavidin (SA)-Biotin Amplification Strategy Based SPR Sensor for Rapid Detection of Escherichia coli

Author's Name:

Institution:

Abstract:

A surface plasmon resonance (SPR) biosensor has been established for rapid detection of Escherichia coli. An activation step performed on CM5 chips was carried using EDC/NHS. The biotin-labeled secondard antibody (goat anti-rabbit IgG) was immobilised onto CM5 chip modified with streptavidin via the biotin-streptavidin interaction. Then, rabbit anti-E. coli ATCC25922 polyclonal antibody (primary antibody) was immobilized on sensor chip by antibody-antigen recognition. The detectable signal was amplified by the amplification of primary and secondary antibodies and the streptavidin-biotin amplification strategy. All the SPR experiments were performed used a Biacore 3000 and CM5 chips. Regeneration was achieved using NaOH in order to detect several samples. The change of RU was linearly correlated with the concentration of E. coli ATCC25922 (r=0.9815). This method exhibited high performance with a dynamic range of $1.5 \times 10 \land 2 \sim 1.5 \times 10 \land 7$ CFU/mL, and a detection limit was $1.5 \times 10 \land 2$ CFU/mL. This method has exhibited great potential in bacteriological testing.

Keywords: SPR sensor; streptavidin-biotin amplification; Escherichia coli ATCC25922; rapid detection

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