

论著

量子点及Cy3免疫荧光染色标记阿尔茨海默病转基因细胞模型APP的比较研究

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摘要:

目的: 对比观察量子点标记的链霉亲和素复合物(QDs-SA)和传统荧光染料Cy3分别标记阿尔茨海默病(Alzheimer's disease, AD)转基因细胞模型淀粉样前体蛋白(APP)的荧光成像性能, 为量子点早期应用于AD的分子影像诊断提供依据。方法: 应用激光共聚焦荧光成像和流式细胞技术, 对QDs-SA量子点以及传统荧光染料Cy3靶向标记AD转基因细胞模型中APP的荧光成像和抗光漂白特性进行检测。结果: 激光共聚焦显微镜下QDs-SA特异标记的APP在胞膜中明显表达, 呈连续分布的橙红色荧光, 细胞膜未见明显的QDs-SA团聚现象。Cy3特异标记APP膜蛋白荧光强度较QDs-SA标记所示的高强度橙红色荧光弱, 且胞膜染色不均匀。QDs-SA量子点的平均荧光密度值(34.2336 ± 4.2455)明显高于传统Cy3荧光染料(21.6023 ± 3.0102 , $P < 0.05$); 488 nm激发光连续激发12 min, QDs-SA量子点荧光标记的APP仍发射较强的橙红色荧光, 荧光强度仅下降27.87%, 明显低于传统染料Cy3的荧光强度下降幅度79.60% ($P < 0.05$)。流式细胞仪检测QDs-SA量子点和Cy3活细胞标记膜蛋白APP阳性率分别为(54.4700 ± 3.4433)%和(54.3800 ± 8.5229)%, 差异无统计学意义 ($P > 0.05$), QDs-SA量子点荧光标记APP的流式荧光图形呈宽端型, 流式平均荧光强度值(1045.4167 ± 47.3623)明显高于Cy3标记的平均荧光强度值(658.5467 ± 55.0591), 差异有统计学意义 ($P < 0.05$)。结论: QDs-SA量子点荧光探针能有效识别AD转基因细胞模型中的APP; QDs-SA量子点标记APP荧光成像在光稳定性和荧光强度等方面均优于传统的Cy3荧光染料标记的免疫荧光成像。

关键词: 阿尔茨海默病 量子点 APP 分子成像

Fluorescence imaging of APP in Alzheimer's disease with quantum dot or Cy3: a comparative study

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Abstract:

Objective To compare the fluorescence intensity and duration of qdots streptavidin conjugate (QDs-SA) with Cy3 as the molecular probe of β -amyloid precursor protein (APP), and to provide evidence for early molecular imaging and diagnosis of Alzheimer's disease (AD). Methods With the help of laser scanning confocal microscope and flow cytometry, the fluorescence probe based on the QDs-SA was used to detect APP in HEK293 cells stably transfected pcDNA3.1/APP, and to compare with conventional fluoroimmunoassay Cy3. Results The immunofluorescence staining detection indicated APP expression was mainly located in the plasma membrane. The mean fluorescence intensity of QDs-SA (34.2336 ± 4.2455) was greater than that of Cy3 (21.6023 ± 3.0102) under the confocal fluorescence microscope ($P < 0.05$). After persistent exciting for 12 min, the fluorescence intensity of APP stained by QDs-SA decreased by 27.87%. The other stained by Cy3 decreased by 79.60%. The positive rate of APP staining had no significant difference between the QDs-SA (54.4700 ± 3.4433)% and Cy3 (54.3800 ± 8.5229)% by flow cytometry, but the mean fluorescence intensity had statistical significance ($P < 0.05$). The QDs-SA (1045.4167 ± 47.3623) was significantly higher than the mean fluorescence intensity of Cy3 (658.5467 ± 55.0591). Conclusion QDs-SA fluorescence probes can effectively recognize APP and are sensitive and exceptionally photostable, suggesting that QDs-SA fluorescence probes could be a potential method in APP detection and offer a novel way for the diagnosis of Alzheimer's disease.

Keywords: Alzheimer's disease; quantum dot; β -amyloid precursor protein; molecular imaging

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