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CEA mRNA转染的成熟树突状细胞体外诱导特异性抗肿瘤作用 点此下载全文

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摘要：

目的：将体外转录的癌胚抗原（carcinoembryonic antigen，CEA）mRNA转染入成熟树突状细胞（dendritic cell，DCs），观察其体外诱导的特异性抗肿瘤作用。方法: GM-CSF、IL-4、TNF-α体外诱导成熟DCs并用流式鉴定。构建pcDNA3.1-CEA载体，体外转录为CEA mRNA，电穿孔法将CEA mRNA转染入DCs。流式细胞术检测转染后DCs中CEA蛋白的表达；MTT法检测DCs刺激T细胞增殖能力；LDH法检测DCs体外诱导的CTL的特异性抗肿瘤作用；ELISA法检测诱导的CTL 上清中IFN-γ的水平。结果: CEA mRNA转染后DCs细胞内CEA蛋白显著高于对照组（83.32% vs 3.34%， P <0.01）。CEA mRNA转染组DCs在效靶比为1∶10时，刺激T细胞增殖作用最强，明显高于未转染组\[(19.11±1.89）% vs （15.59±0.70）%， P <0 05\]。CEA mRNA转染组DCs能产生CEA特异性的CTL效应，在效靶比为5∶1、10∶1、20∶1和40∶1时杀伤率分别为\[（ 5.42± 0.87）%、（14.09±1.13）%、（27.16±0.72）%、（32.49±0.84）%， P <0.01\]，而未转染组和对照靶细胞均无杀伤作用。转染组DCs诱导的CTL上清中IFN-γ分泌量显著高于未转染组\[（141.73±28.61） vs （9.45±4.63）pg/ml， P <0 01\]。结论：CEA mRNA转染的成熟DCs体外能产生特异性抗肿瘤作用，为研制CEA RNA-DCs疫苗提供了实验依据。

关键词：树突状细胞 癌胚抗原 RNA 疫苗 免疫治疗

Transfection of mature dendritic cells with carcinoembryonic antigen mRNA induces specific anti-tumor effects in vitro Download Fulltext

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Abstract:

Objective:To transfect transcripted-carcinoembryonic antigen (CEA) mRNA into mature dendritic cells (DCs) in vitro , so as to observe the specific anti-tumor effect of DCs. Methods: Mature DCs was induced by GM-CSF, IL-4 TNF-α and then identified by FACS. pcDNA3.1-CEA plasmid was constructed and transcripted to CEA mRNA in vitro , and CEA mRNA was then tranfected into mature DCs by electroporation; CEA protein expression in DCs was examined by FACS, the proliferation of T cells induced by DCs was examined by MTT, the ability of DCs to induce specific anti- tumor responses of CTL was examined by LDH, and the level of IFN-γ in CTL supernatant was determined by ELISA. Results: CEA protein in CEA mRNA tranfected DCs was significantly increased compared with that in the control group (83.32% vs 3.34%, P <0.01). CEA mRNA tranfected DCs showed a stronger ability to induce the proliferation of T cells compared with control DCs when the ratio of effect cell to target cell (E∶T) being 1∶10 (\[19.11±1.89\]% vs \[15 59±0.70\]%, P <0.05). CEA mRNA tranfected DCs induced specific anti-tumor responses of CTL, the cytotoxic rates being (5.42±0.87)%, (14.09±1.13)%, (27.16±0.72)%, and (32.49±0.84)% (P <0.01) when the E∶T were 5∶1, 10∶1, 20∶1 and 40∶1, and untransfected DCs and control target cells showed no cytotoxic effects. The level of IFN-γ in CTL supernatant induced by CEA mRNA tranfected DCs was significantly increased compared with that in the untransfected group (\[141.73±28.61\] pg/ml vs \[9.45±4.63\] pg/ml, P <0.01). Conclusion:Mature DCs transfected with CEA mRNA can induce specific anti-tumor responses, which provides a theoretical basis for CEA RNA-DCs vaccine.

Keywords:dendritic cell carcinoembryonic antigen RNA vaccine immunotherapy

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