

巢式RT-PCR 扩增CK20、MMP-7 诊断大肠癌淋巴结微转移的初步研究

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Study on Detecting Colorectal Cancer Micrometastases in Lymph Nodes by Nested Reverse Transcriptase-Polymerase Chain Reaction Amplification of Cytokeratin 20 and Matrix Metalloproteinase-7

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摘要

目的 建立巢式逆转录聚合酶链式反应(nested reverse transcriptase-polymerase chain reaction, nested RT-PCR)扩增CK20mRNA、MMP-7mRNA, 检测大肠癌淋巴结微转移。方法 据CK20、MMP-7基因序列设计四对引物, 建立巢式RT-PCR扩增体系, 检测CK20 mRNA、MMP-7 mRNA在30例行根治性手术大肠癌患者187个淋巴结中及10例良性肠道疾病患者25个淋巴结中的表达, 并与传统组织学检查相比较。结果 大肠癌患者187个淋巴结中75个CK20 mRNA表达, 阳性率40. 11%(75 / 187), 97个MMP-7 mRNA表达, 阳性率51. 87%(97 / 187), 61个CK20 mRNA及MMP-7 mRNA表达, 阳性率32. 62%, 病理组织学检查仅57个淋巴结有肿瘤转移; 10例良性肠道疾病25个淋巴结扩增结果阴性。结论 巢式逆转录聚合酶链式反应检测淋巴结CK20 mRNA、MMP-7 mRNA的表达是诊断大肠癌微细胞转移灵敏特异的指标, 其结果较常规病理组织学检查敏感, 可对精确大肠癌的临床分期、判断患者预后及治疗提供一定理论依据。

关键词: 结肠直肠癌/ 病理学 逆转录聚合酶链式反应 角蛋白/ 生物碱 淋巴转移

Abstract: Objective To develop a nested reverse transcriptase polymerase chain reaction method to amplify Cytokeratin 20mRNA and Matrix Metalloproteinase-7mRNA for detecting colorectal cancer micro-metastases in lymph nodes. Methods The RT-PCR system was set up by using nest primer. The presence of CK20 mRNA and MMP-7 mRNA was investigated by nested RT-PCR in 100 lymph nodes from 30 colorectal cancer patients who underwent curative operation. Twenty-five lymph nodes from 10 patients with benign intestinal disease were served as negative control. The results of nested RT-PCR were compared with ones of routine histology. Results The positive rate of CK20 mRNA and MMP-7 mRNA in 187 lymph nodes from 30 colorectal cancer patients was 40. 11%(75/ 187), 51. 87%(97/ 187) respectively, while only 57 samples were histologically positive. The expression of CK20 mRNA and MMP-7 mRNA in 25 lymph nodes of 10 cases of control group were all negative. Conclusion Nested reverse transcriptase polymerase chain reaction was a sensitive and specific method to indicate the expression of CK20 mRNA and MMP-7 mRNA in lymph nodes as a helpful marker of micrometastases of colorectal carcinoma cells. It is superior to routine histopathological assay in examining lymph node metastases. The detection of CK20 mRNA and MMP-7 mRNA in lymph nodes may contribute to improve the accuracy of clinical staging, determine patients' prognosis and provide information for adjuvant therapy.

Key words: Colorectal neoplasms/ pathology Reverse transcriptase polymerase chain reaction Keratin/ biosynthesis Lymphatic metastasis

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