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论著

肝癌相关基因的分子克隆和可变剪切分析

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摘要: 目的: 获取肝癌相关基因(hepatoma associated gene, HTA)的mRNA分子全长序列并对其 可变剪接进行分析,检测其转录本在各肝癌细胞系中的表达,为进一步研究该基因在肝癌发生、发展 中的作用奠定基础。方法: 用3'RACE 和5'RACE方法扩增HTA基因的全长序列并对其序列信息和可变 剪接进行扩增和测序分析,用Northern印迹检测该基因转录本在不同肝癌和正常肝细胞系中的表达。 结果: HTA基因全长序列为1414 bp, 经分析该序列包含3个外显子, 2个内含子, 其中2号内含子在可 ▶把本文推荐给朋友 变剪接中被作为外显子表达。Northern印迹显示HTA基因1.7 kb转录本和1.4 kb转录本均表达于恶性 肝癌细胞系,而不表达于正常细胞系。结论:成功获得了HTA基因的全长序列并对其可变剪接进行了 分析,2个大小不同的转录本均在肝癌细胞系中特异性表达,作为一个肝癌相关基因值得进一步深入 研究。

关键词: 肝癌相关基因 全长克隆 可变剪接

Molecular cloning and alternative splicing analysis of hepatoma associated gene HTA

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Abstract: Objective: To obtain the full length cDNA sequences of hepatoma associated gene HTA, analyze its alternative splicing, detect the expression pattern of 2 HTA gene transcripts in different hepatic cell lines, and to establish a base for further study of HTA gene function in hepatocellular carcinoma (HCC) occurence and development. Methods: The full length cDNA of HTA gene was cloned by rapid amplification of cDNA 3' ends (3'-RACE), rapid amplification of cDNA 5' ends (5'-RACE) and DNA sequencing. The gene structure and alternative splicing were analysed. Northern blot assay was performed to detect the expression pattern of 2 HTA gene transcripts in different hepatic cell lines. Results: The full length of HTA gene was 1414 bp, composed of 3 exons and 2 introns, and the second intron could be retained in mRNA. Northern blot assay showed that 2 transcripts of HTA mRNA(1.4 kb and 1.7 kb) could express in the HCC cell lines HepG2 and QGY-7703, but not in the non-malignant cell line L-02 and HUVEC. The expression level of 1.4 kb transcript was much higher than 1.7 kb one.

Conclusion: This study successfully has obtained the full length cDNA of HTA gene, and analysed the gene sequence and alternative splicing, 2 transcripts of HTA mRNA specifically expressed in HCC cell lines. As a hepatoma associated gene, HTA deserves further investigation.

Keywords: HTA full length cloning alternative splicing

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