# 常氏肝癌细胞cDNA文库的构建及ADAMs相关基因的免疫筛选与序列 分析

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摘要 抽提常氏肝癌细胞的总RNA,以oligo(dT)为引物,通过两次转换模板,采用LD-PCR合成全长cDNA,在cDNA 两端引入SfiI的酶切位点。以λTriplEX2为载体经过包装后构建了常氏肝癌细胞cDNA表达文库。以ADAMs通用抗体,用免疫筛选技术从常氏肝癌cDNA文库中筛选出22个阳性克隆,经测序分析和BLAST检索,证实其中一个为新基因,部分区域具有蛋白酶的功能,对该基因进行了GenBank登录,获注册号为AY078070。该基因的克隆为研究ADAMs相关基因的功能奠定了基础。

关键词 常氏肝癌细胞 cDNA文库 ADAMs抗体 免疫筛选 基因克隆 同源检索 分类号

# **Construction of Chang Liver cDNA Library and Immunoscreening of ADAMS Related Genes**

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#### Abstract

Total RNA was isolated from Chang Liver cell. The full-length cDNA was synthesized using LD PCR by RNA  $5 < \sup >$ ,  $</ \sup >$ ; end switching mechanism, oligo(dT) as primer. Sfil digestion sites were introduced at both  $3 < \sup >$ ,  $</ \sup >$ ; and  $5 < \sup >$ ,  $</ \sup >$ ; end of cDNA. The cDNA was ligated with  $\lambda$ TriplEX2 vector and packaged. Twenty-two positive clones were obtained by immunoscreening to the library, they were sequenced and blast in NCBI, one of them was identified a novel gene. It was submitted to GenBank and obtained accession nubmer AY078070. It may lay down a foundation for studying on functions of ADAMs related gene.

**Key words** Chang Liver cDNA Library ADAMs antibody immunoscreening gene cloning BLAST

DOI:

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