研究报告

戊型肝炎病毒第4基因型中国株ORF2编码蛋白 的抗原表位分析

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收稿日期 2006-11-19 修回日期 2007-1-15 网络版发布日期 2007-4-12 接受日期

以戊型肝炎病毒(HEV)第4基因型中国株ORF2编码蛋白p166Chn制备单克隆抗体(McAbs),同时制备20个N端或C端逐 渐截短的p166Chn截短蛋白,与7种不同基因型和亚型的p166蛋白一起,通过ELISA、免疫印迹(Western blot)以及 竞争抑制实验对主要存在于我国的HEV第4基因型毒株进行抗原表位分析。结果发现所制备的McAbs与p166Chn截短 蛋白的免疫反应有两种类型,以1G10为代表的McAbs能与N端不短于aa477、C端不短于aa613的截短蛋白反应,其针<mark>▶浏览反馈信息</mark> 对的抗原表位是构象依赖型表位,依赖于aa477~aa613肽链区段;而McAb 2F11则能与N端不短于aa474、C端不短于 aa617的截短蛋白反应,其针对的抗原表位也是构象表位,但需依赖于较长的肽链区段(aa474~aa617)。竞争抑制 实验显示两类McAbs互不抑制,进一步证实了所发现的两个抗原表位在空间位置上的不同。更有意义的是,两类 McAbs均能与其他不同HEV基因型和亚型来源的p166重组蛋白发生阳性反应,表明这两个抗原表位是HEV基因型共同 相关文章 性的,可以在世界各国分布的不同基因型HEV毒株中诱导交叉免疫。

关键词 戊型肝炎病毒 单克隆抗体 基因型 表位 分类号

Characterization of antigenic epitopes of ORF2 encoded proteins of hepatitis E virus genotype 4 identified in China

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Abstract

<P>To characterize antigenic epitopes of hepatitis E virus (HEV) genotype 4 that was first identified in China a few years ago, a recombinant protein, p166Chn, encoded by HEV genotype 4 ORF2 was used to prepare anti-p166Chn McAbs. Simultaneously, twenty N- or C-terminal truncated p166Chn proteins were generated. Immunoreactivity between the McAbs and the truncated proteins as well as seven p166 recombinant proteins derived from different HEV genotypes and subgenotypes was detected by indirect ELISA, Western blot and competition inhibition ELISA. Two reactive profiles were observed with different McAbs and different truncated proteins. The McAbs, represented by 1G10, reacted with those N-terminal truncated proteins beginning at upstream of aa477 and those Cterminal truncated proteins ending at down-stream of aa613, suggesting that the epitope recognized by 1G10 relied on the region of aa477-aa613 and was conforma-tion-dependent. While McAb 2F11 was reactive to those truncated p166Chn proteins beginning at upstream of aa474 or ending at downstream of aa617, indicating that the epitope recognized by 2F11 was also conformationdependent and relied on a longer peptide of aa474-aa617. However, the two

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groups of McAbs didn't inhibit each other when tested by a compe-tition inhibition ELISA, which confirmed the different spatial positions of the two epitopes. Furthermore, when p166 pro-teins derived from different HEV genotypes and subtypes were applied, all of the McAbs prepared against pChn166 of genotype 4 identified in China could react with the proteins of genotype 1, 2 and 3 distributed worldwide. The data sug-gested that the two identified epitopes were HEV genotype-common and played significant effects on cross immunoreactiv-ity between different HEV genotypes.

Key words Hepatitis E virus monoclonal antibodies genotype epitope

DOI: 10.1360/yc-007-0637

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