

大鼠热激因子结合蛋白1全长cDNA克隆与分析Cloning and Analysis of Rat Heat Shock Factor Binding Protein 1 cDNA

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摘要

热激因子结合蛋白1 (heat shock factor binding protein 1, HSBP1) 是一种新发现的高保守、低分子量、定位于核中的一种转录因子, 它可抑制HSF1的转录域活性, 并协同HSP70负调控热激反应。在哺乳动物中仅有人与小鼠的HSBP1cDNA序列被克隆, 大鼠中的HSBP1尚未有人报道, 我们根据人与小鼠的HSBP1氨基酸的N端和C端保守序列设计了简并引物, 采用RT-PCR方法从大鼠神经胶质瘤细胞株中克隆大鼠HSBP1的片段, 然后采用Southern 印迹方法从建立的神经胶质瘤细胞的cDNA文库中调取了它的cDNA全长序列, 递交给GenBank, 获登陆号为AF522937。并借助于大鼠基因组测序成果, 将大鼠HSBP1基因定位于19q12, 发现HSBP1基因有3个内含子和4个外显子, 其中外显子1和外显子4之间距离5829bp。且对HSBP1的Unigene检索显示HSBP1广泛存在于大鼠各组织、器官中, 揭示HSBP1在生理活动中起着非常重要的作用。本文并根据已发表的其他物种的HSBP1的氨基酸序列用DNAMAN 软件作了它的同源关系分析, 结果显示进化关系较近的物种中, HSBP1氨基酸序列的相似性与其从形态解剖所得的系统进化关系是一致的。

Abstract: Heat shock factor binding protein 1 (HSBP1) is a nuclear-localized, novel, conserved, low molecular weight (<100 residues) transcriptional factor, which may repress the activity of the heat shock factor 1 (HSF1) by binding HSF1 active trimerization domain. HSBP1 gene have been cloned in human and mouse, but not reported in rat. In this paper, a pair of consensus degenerate primers were designed based on N-terminal and C-terminal conservative amino acid sequence. Using RT-PCR method, hsbp1 gene fragment was amplified and cloned from total RNA extracted from rat C6 glioma cells. Then the EST was probed to isolate the rat full-length hsbp1 cDNA by in situ hybridization from a rat C6 glioma cells cDNA library. The full-length hsbp1 was deposited in GenBank (accession NO. AY522937). It was blasted in RGD (rat genome database) and was localized in 19q12 and composed of four exons and three introns. The distance between the first exon and the fourth exon was 5829bp. Then its Unigene was searched, results showed HSBP1 existed widely in all kinds of organs and tissues, the data suggested that it may play a important roles in physiological activity. In addition, the sequence similarity and phylogenetic relationship were compared with DNAMAN tool. The result showed the relationship is consistent between the similarity of amino acid sequence and phylogenetic evolution from morphological of those species which were nearly in evolution.

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