

CLONING AND IDENTIFICATION OF A CONSTITUTIVELY EXPRESSED SPLICING ISOFORM OF *ADAR1* (*CIADARIA*) IN

Ctenopharyngodon idella

Xiancheng Liu, Huiling Mao[§], Chengyu Hu[§]

Department of Bioscience, College of Life Science, Nanchang University, Nanchang 330031, China

ABSTRACT

Belonging to ADAR (adenosine deaminase acting on RNA) family, ADAR1 catalyzes the deamination of adenosine to inosine within dsRNA. In mammal, there are two well known splicing types, i.e. the interferon (IFN) inducible ~150 KD protein (ADAR1-p150) and the constitutively expressed ~110 KD protein with N-terminally truncated (ADAR1-p110). In our previous study, we have cloned the complete genome of grass carp *ADAR1* (*CiADAR1*). In this study, we identified a new constitutively expressed splicing isoform gene, *CiADAR1a* (*CiADAR1a*). The complete genome of *CiADAR1a* has 15 exons and 14 introns. Different from that of *CiADAR1*, the transcriptional start site of *CiADAR1a* promoter is mapped within a truncate exon 2. Its full-length cDNA is comprised of a 5'UTR (359 bp), a 3'UTR (229 bp) and a 2952 bp ORF encoding a polypeptide of 983 amino acids. *CiADAR1a* contains one Z-DNA binding domain, three dsRNA binding motifs and a highly conserved hydrolytic deamination domain. Western blot showed that ADAR1a was constitutively expressed and did not change after stimulation with poly(I:C) in CIK cells. To further confirm whether the promoter activity is affected by IRF, we cloned *CiADAR1a* promoter sequence. *CiADAR1a* promoter is 1038 bp in length containing 4 IRF-E. *In vivo*, co-transfection of pcDNA3.1-IRF1 (and pcDNA3.1-IRF3 respectively) with pGL3-*CiADAR1a* promoter into CIK cells, the Dual luciferase activity did not change under *CiIRF1* and *CiIRF3* treatment respectively. It revealed that *CiADAR1a* is a constitutively expressed protein *in vivo*.

Keywords

Adenosine demination; ADAR1; IFN; RNA editing; Transcriptional regulation

[§]Corresponding author. Tel: +86-791-8396-9530

E-mail address: 13507085896@163.com (H. Mao); hucy2008@163.com (C. Hu)