

## 草鱼 $\alpha$ -淀粉酶基因5'侧翼序列克隆与分析

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### Cloning and analysis of 5' flanking sequence of $\alpha$ -amylase gene from *Ctenopharyngodon idellus*

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#### 摘要

应用PCR和Genome Walking技术克隆获得长度为168 bp的草鱼 (*Ctenopharyngodon idellus*)  $\alpha$ -淀粉酶基因外显子 I 序列和2 063 bp的5'侧翼序列。将草鱼 $\alpha$ -淀粉酶基因外显子 I 序列与已知几种鱼类 $\alpha$ -淀粉酶基因外显子 I 序列比对, 相似度为68%~86%。将草鱼 $\alpha$ -淀粉酶基因5'侧翼序列进行生物信息学分析, 确定了其转录起始区域及转录起始位点(TSS); 在TSS上游30 bp处有1个TATA-box, -58 bp处有CCAAT-box; 在5'侧翼序列中还发现有GATA-1、OCT-1、GR、HNF-3、AP1和SP1等转录因子结合位点。草鱼 $\alpha$ -淀粉酶基因5'侧翼序列的克隆, 为进一步研究不同食性鱼类 $\alpha$ -淀粉酶基因侧翼序列的差异、鱼类 $\alpha$ -淀粉酶基因的表达、功能及调控机理奠定基础。

**关键词:** 草鱼,  $\alpha$ -淀粉酶基因, Genome Walking, 5'侧翼

#### Abstract:

PCR and Genome Walking methods were applied to obtain a 168 bp  $\alpha$ -amylase gene exon I sequence and a 2 063 bp  $\alpha$ -amylase gene 5' flanking sequence from grass carp (*Ctenopharyngodon idellus*). The sequence alignment of  $\alpha$ -amylase gene exon I between grass carp and other fishes showed that the similarity was 68%~86%. The sequence analysis of  $\alpha$ -amylase gene 5' flanking sequence of grass carp confirmed a transcription initiation region and a transcriptional start site (TSS). There were a TATA-box at 30 bp and a CAAT-box at -58 bp. There were some transcriptional factor binding sites in the  $\alpha$ -amylase gene 5' flanking sequence, such as GATA-1, OCT-1, GR, HNF-3, AP1 and SP1. The  $\alpha$ -amylase gene 5' flanking sequence of grass carp was cloned successfully, which provides references for studies on differences of  $\alpha$ -amylase gene 5' flanking sequence among different feeding fishes, as well as on the expression, function and regulation mechanism of  $\alpha$ -amylase gene in fishes.

**Key words:** *Ctenopharyngodon idellus*  $\alpha$ -amylase gene Genome Walking 5' flanking sequence

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