

绵羊ILK基因的克隆及其在毛囊生长期的表达

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摘要 整合素连接激酶(ILK)是一种支架蛋白, 在毛囊发育过程中发挥重要作用。文章利用PCR技术, 首次获得绵羊ILK基因的编码区全长序列, 并进行了生物信息学分析; 同时对该基因的组织表达谱及其在不同绵羊品种毛囊生长期皮肤组织中的表达变化进行了研究。结果表明, 绵羊ILK基因ORF全长1 359 bp, 编码452个氨基酸。ILK蛋白结构经预测含有3个锚定重复序列和1个激酶结构域, 并存在多个磷酸化位点和蛋白激酶C的磷酸化位点。半定量RT-PCR结果显示该基因在绵羊心脏、肝脏、脾脏、肺脏、肾脏、骨骼肌、皮肤和小肠组织中均有表达, 皮肤、脾脏和肝脏中表达量较高; 实时荧光定量PCR结果表明在3~5月份(毛囊生长起始期), ILK基因在中国美利奴超细型和哈萨克羊皮肤组织中的表达水平较高并均呈逐月上升趋势; 在6~10月份(毛囊生长期), 中国美利奴超细型皮肤ILK基因表达水平高于同期的哈萨克羊。分析认为ILK可能在调控绵羊次级毛囊生长发育过程中起一定作用。

关键词: 整合素连接激酶 基因克隆 毛囊 绵羊

Abstract: Integrin linked kinase (ILK) is a scaffold protein, which plays important roles in hair follicle development. The cDNA sequence of novel ILK gene in sheep was cloned by PCR method and analyzed by bioinformatics. Tissue expression profiling in eight tissues and temporal profiling at different wool follicle anagen stages in skin was analyzed. The results showed that the whole open reading frame (ORF) of ILK gene was 1 359 bp in length, which encoded 452 amino acids. Bioinformatic analysis indicated that the secondary structure of ILK gene was mainly made up of three ankyrin repeats and a kinase domain, and there were multiple phosphorylation and Protein Kinase C sites in this gene. The RT-PCR result confirmed that ILK mRNA was expressed in heart, liver, spleen, lung, skeletal muscle, skin, and small intestine, and the expression level was much higher in skin, spleen, and liver than others. The q-PCR analysis demonstrated that the expression level of ILK was significantly increased from March to May (early follicle anagen initiation) in both sheep breeds, Chinese Merino and Kazakh sheep, and there were certain differences from June to October between the two breeds. The above results indicated that ILK gene may play key roles in regulating secondary follicle growth.

Keywords: integrin-linked kinase, gene cloning, hair follicle, sheep

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