



### 刚地弓形虫ROP11基因的克隆及生物信息学分析

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#### Cloning and Bioinformatics Analysis of Rhoptyry Protein 11 of *Toxoplasma gondii*

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摘要

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**摘要** 提取刚地弓形虫 (*Toxoplasma gondii*) RH株速殖子总RNA, 根据棒状体蛋白11 (ROP11) 全长编码序列 (登录号为DQ077905) 的开放阅读框设计引物并进行逆转录PCR (RT-PCR) 扩增, PCR产物经 *EcoR* I 和 *Not* I 酶切后与原核表达载体pGEX-6P-2连接, 重组质粒转化大肠埃希菌 (*E. coli*) XL-Blue, 阳性菌落经PCR和双酶切鉴定, 并测序。对所得序列进行生物信息学分析。结果显示, RT-PCR扩增产物约为1500 bp。菌落PCR及双酶切结果正确。测序结果显示, 获得的ROP11基因片段为1548 bp (登录号为KC456639), 与GenBank上已有的弓形虫ROP11序列相比, 序列一致性为99%。生物信息学分析发现, ROP11编码蛋白质的预期相对分子质量为 $M_r$  57 020, 包括有12个保守结构区域, 其前26个氨基酸残基构成信号肽, 丝氨酸/苏氨酸蛋白激酶催化区域位于170~511氨基酸, 且有2个潜在的N-糖基化位点。

**关键词:** 刚地弓形虫 棒状体蛋白11 原核表达 生物信息学分析

**Abstract:** Total RNA was extracted from tachyzoites of RH strain of *Toxoplasma gondii*. The open reading frame of ROP11 gene was amplified by using a pair of specific primers designed according to the coding sequence of ROP11 gene (Accession No. DQ077905). The RT-PCR product was digested by restriction enzyme *EcoR* I and *Not* I, and then ligated into a pGEX-6P-2 vector. The recombinant plasmid was transferred into *E. coli* XL-Blue. The positive clones was selected by colony PCR, and confirmed by the double restriction enzyme digestion and sequencing. The RT-PCR product was 1548 bp. The recombinant plasmid was confirmed by colony PCR and double restriction enzyme digestion. Sequencing results showed that the obtained ROP11 gene was 1548 bp (Accession No. KC456639). There was a high sequence consistency (99%) between the obtained ROP11 gene sequence and the *Toxoplasma* ROP11 gene from GenBank. Bioinformatics analysis showed that the ROP11 protein ( $M_r$  57 020) consisted of the signal peptide (amino acids 1-26), 12 conservative domains, a serine/threonine protein kinase catalytic domain (amino acids 170-511), and two potential N-glycosylation sites.

**Keywords:** *Toxoplasma gondii* Rhoptyry protein 11 Prokaryotic expression Bioinformatics analysis

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