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863课题进展

仿刺参cytb和β-actin基因表达稳定性比较

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摘要:

基因表达分析需要采用内参基因来校正目的基因的表达量。采用半定量RT-PCR的方法分析了cytb和β-actin基因在仿刺参(Apostichopus japonicus)未受精卵、受精卵、多细胞期、囊胚期、原肠期、小耳状幼体、中耳状幼体、大耳状幼体、樽型幼体、五触手幼体、稚参11个发育阶段和幼参的体壁、体腔细胞、肠道和呼吸树中的表达情况。结果表明:cytb在不同发育阶段和不同组织中稳定表达;β-actin基因在稚参之前不同发育阶段中表达水平有显著差异,在幼参的体壁、肠道和呼吸树中稳定表达。此外,cytb在lipopolysaccgarides (LPS)刺激前后的原肠胚、小耳状幼体、中耳状幼体、大耳状幼体、樽型幼体、稚参和幼参四种组织中表达量无显著差异;β-actin基因在LPS刺激前后的幼参体腔细胞、肠道和呼吸树中表达量无显著差异。本研究为仿刺参功能基因表达分析中,cytb与β-actin基因作为内参基因的可行性提供了依据。

关键词: 仿刺参;cytb基因;β-actin基因;LPS刺激;内参基因

Stability Comparison of cytb and β-actin Genes Expression in Sea Cucumber (Apostichopus japonicus)

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Abstract:

Analysis of gene expression commonly uses internal control gene for normalization. The mRNA levels for cytb and β-actin genes were detected by using RT-PCR at 11 larval development stages of sea cucumber（Apostichopus japonicus）including unfertilized egg, fertilized egg, cellulous stages, blastula, gastrula, early auricularia, auricularia, late auricularia, doliolaria, pentactula, juvenile and in different tissues of young sea cucumbers including body walls, coelomocytes, intestines and respiratory trees. The results showed that cytb gene expression was stable at all development stages and in the four tissues. The mRNA levels for β-actin gene showed significant differences at different development stages, but were stable in the tissues of body walls, intestines and respiratory trees. Moreover, cytb mRNA showed no significant changes in expression after a LPS challenge at the stages of gastrula, early auricularia, auricularia, late auricularia, doliolaria and juvenile or in the four tissues of young sea cucumbers. The expression of β-actin gene was also stable in the tissues of coelomocytes, intestines and respiratory trees after a LPS challenge. This study provides information for the selection of cytb gene and β-actin gene as suitable internal control gene in target genes expressions research of sea cucumber.

Keywords: sea cucumber(Apostichopus japonicus);cytb gene;β-actin gene;LPS challenge;internal control gene

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