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微信公众号: 大豆科学

[1]李飞武,李葱葱,邢珍娟,等.第二代抗草甘膦大豆PCR检测方法研究[J].大豆科学,2009,28(02):296-300.
 [doi:10.11861/j.issn.1000-9841.2009.02.0296]
 LI Fei-wu,LI Cong-cong,XING Zhen-juan,et al.Establishment of an Event-specific Qualitative PCR Method for Detection of Roundup RReady2YieldTM[J].Soybean Science,2009,28(02):296-300.[doi:10.11861/j.issn.1000-9841.2009.02.0296]

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第二代抗草甘膦大豆PCR检测方法研究

《大豆科学》 [ISSN:1000-9841 /CN:23-1227/S] 卷: 第28卷 期数: 2009年02期 页码: 296-300 栏目:
 出版日期: 2009-04-25

Title: Establishment of an Event-specific Qualitative PCR Method for Detection of Roundup RReady2Yield™
 文章编号: 1000-9841(2009)02-0296-05
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 关键词: 转基因生物 (KeySearch.aspx?type=Keyword&Sel=转基因生物); Roundup RReady2Yield™ (KeySearch.aspx?type=Keyword&Sel=Roundup RReady2YieldTM); 转化体特异性PCR (KeySearch.aspx?type=Keyword&Sel=转化体特异性PCR)
 Keywords: Genetically modified organism (KeySearch.aspx?type=Keyword&Sel=Genetically modified organism); Roundup RReady2Yield™ (KeySearch.aspx?type=Keyword&Sel=Roundup RReady2YieldTM); Event-specific PCR (KeySearch.aspx?type=Keyword&Sel=Event-specific PCR)
 分类号: S565.1
 DOI: 10.11861/j.issn.1000-9841.2009.02.0296 (http://dx.doi.org/10.11861/j.issn.1000-9841.2009.02.0296)
 文献标志码: A

摘要: 为建立转基因大豆Roundup RReady2Yield™(RR2Y)转化体特异性定性PCR检测方法,以lectin基因作为内参基因,根据RR2Y外源插入片段5'端与植物基因连接区序列设计特异性引物,从RR2Y中特异性地扩增出223bp的预期产物。对该方法进行重现性、特异性、灵敏度、稳定性和可重复性测试,结果显示:该方法能够特异性地检测出RR2Y转化体;将100% RR2Y基因组DNA用A3244基因组DNA进行梯度稀释,以100 ng DNA为模板,该方法的检测灵敏度达到0.05%,约为40个起始模板拷贝;以RR2Y DNA含量为10%、1%、0.1%的样品为模板,进行稳定性和可重复性,假阴性率为0。结果表明:此方法适用于RR2Y的转化体特异性定性PCR检测。

Abstract: Roundup RReady2Yield™(RR2Y) is a glyphosate-tolerant genetically modified soybean, which is approved to be imported and used as raw material in China. For labeling them according to the regulations on safety of agricultural genetically modified organisms, an event-specific qualitative PCR method for RR2Y is urgently needed to be established, which is the aim of this study. The specific primers were designed based on the 5' - junction sequences of the exogenous integrant of RR2Y, and amplification products of 223 bp were obtained. The ruggedness, specificity, sensitivity, stability and repeatability of this method were tested. The results showed that RR2Y event can be distinguished specifically from other GM and non-GM crops by using this method, and the limit of detection is up to 0.05%.

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备注/Memo 基金项目: 转基因生物新品种培育重大专项资助项目(2008ZX08012-001)。

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更新日期/Last Update: 2014-10-04