

Identification of genetically modified organisms by detection of target gene pattern using DNA microarrays

(Received January 6, 2014)

(Accepted June 24, 2015)

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Abstract

For the identification and quantification of genetically modified organisms (GMOs), one possible simple alternative method relies on the detection of DNA fragments synthesized using random primers without the need for nucleic acid amplification (for example, PCR) on DNA microarrays. Here, we tested simple detection protocols with a DNA microarray, and consequently, we were able to identify five selected GM maize lines by the pattern of spots detected on the DNA microarray. Our protocol requires no specific primers in the target DNA synthesis steps; all target DNA is synthesized by random 9-mer primers in one tube. This study suggests the possibility of detecting transgenes in GMOs and identifying GMO lines by the pattern of independently detected spots, irrespective of the position of the target gene sequences in the genomic DNA of each GMO line.

Keywords : DNA microarray, genetically modified organism (GMO), screening detection, identification

I Introduction

Statistical data on the worldwide area under cultivation with genetically modified organisms (GMOs) showed that this area was less than five hectares in 1996, but has been increasing, and the acreage of GMOs was over 181,500,000 hectares in 2014¹⁾. In Japan, the year 1996 was memorable because the first three GMOs were introduced with genes for herbicide and harmful insect resistance, traits allowed as being safe for food use according to Japanese guidelines for the safety assessment of foods derived from plants containing recombinant DNA. Since then, new GMOs have been commercially developed, and more than 300 GMOs have been authorized through a safety assessment for commercial use in Japan as of May 2015. In particular, in maize, two or more individual genetically modified (GM) plants having different traits such as herbicide and harmful insect resistance have been hybridized using conventional breeding, and many varieties of hybridized

GMOs, called stacked GMOs, have been produced. As of May 11, 2015, the Japanese Ministry of Health, Labour and Welfare reported that 28 GM maize varieties produced by a single-gene introduction event were authorized as safe food; 273 stacked varieties hybridized in combination with these 28 GM maize varieties have passed safety assessment in Japan. The number of such stacked GMOs having two or more transgenes derived from hybridization of single-event GM varieties has increased, and the genetic structure of stacked GMOs is getting more complex as a result of multiple rounds of hybridization of single-event GM varieties²⁾.

Only GMOs authorized by a food and feed safety assessment can be imported and commercially distributed in most countries. In order to manage the risk regulation of GMOs, and to accept authorized ones but prohibit unauthorized ones on the market³⁾, detection methods have been required to identify individual GMOs. PCR is mainly used to detect GMOs because their nucleic acid sequences often remain in

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