Research letter

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Verifying correlation between qualitative and quantitative PCR in genetically modified soybean using real-time PCR

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Abstract

The Notice of the Deputy Commissioner of the Consumer Affairs Agency dated 15th September 2021 (No. 389) regarding “Partial Amendments to ‘Food Labeling Standards’” (24th amendment), which is a notice amending the Notice of the Deputy Commissioner of the Consumer Affairs Agency dated 30th March 2015 (No. 139) regarding “Food Labeling Standards”, added a new inspection method for soybean grains (to determine contamination with genetically modified agricultural products) to the “Attachment: Inspection methods for genetically modified foods that have been safety reviewed.” We verified the correlation between qualitative PCR analysis using the ΔΔCq method and quantitative PCR assessment that has been traditionally used as the test method for determining appropriateness of food labeling of identity preserved handling. Qualitative PCR was performed on six soybean samples (samples 2–7) collected in 2021 from businesses in Mie Prefecture that import from countries outside Japan and one sample of domestically produced soybeans (sample 1) that was labelled as non-genetically modified; additionally, the RRS, LLS, and RR2 contamination rates were known for samples 2-7, which had passed quantitative PCR testing before qualitative PCR was performed. In these samples, quantitative PCR result for sample 7 showed that RRS, LLS, and RR2 contents were 0.02%, 0.00%, and 0.13%, respectively. Quantitative PCR result for sample 4 showed that RRS, LLS, and RR2 contents were 0.00%, 0.00%, and 0.04%, respectively. In qualitative PCR, sample 7 was assessed as positive, whereas sample 4 was assessed as negative. These results suggest that the likelihood of a positive result in qualitative PCR increases when the RR2 content alone exceeds 0.13%. Moreover, the qualitative PCR result may be positive even if the quantitative PCR result is below the limit of quantification (0.3%).

Keywords : genetically modified (GM), ΔΔCq method, RRS, LLS, RR2

Ⅰ Introduction

Genetic modification of crops enhances desirable traits such as herbicide resistance, bacterial or insect resistance, and nutritional value[1]. However, consumers were growing increasingly concerned regarding genetically modified foods worldwide. Nevertheless, the commercial adoption of genetically modified foods is also increasing; for example, the cultivation of genetically modified soybeans in the United States has increased from 7.4% in 1996 to 75% in 2002[2], and soy protein is widely used in processed foods in developed countries[3]. In contrast, data from the fourth Eurobarometer survey conducted in 1999 showed that although public attitudes were positive toward genetic modification in medical and environmental applications, opposition to genetically modified foods increased[4].

A Japanese survey on the public perception of genetically modified crops showed strong resistance to genetically modified crops owing to safety concerns[5]. Furthermore, concerns also raised about the detrimental effects of biotechnology on fruit and vegetables[6]. To appease the concerns regarding the safety of genetically modified foods, the US Food and Drug Administration implemented measures such as holding public meetings to hear criticism of genetically modified foods[7] On the other hand, some reports indicate that negative impressions of GM technology are on the decline according

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