

Short communication

Comparative evaluation of DNA extraction methods for food crops

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Introduction

In recent years, global distribution of transgenic plants has generated a dramatic increase of the presence of genetically modified organisms (GMOs) in seeds, grains, food and feed. The efficient system for monitoring of GMOs needs reliable and efficient methods for detecting plant species and GMOs in food. DNA is the preferred analyte for seeds, raw materials, ingredients and processed foodstuffs as it is a rather stable molecule. Because of its high specificity and sensitivity, DNAbased polymerase chain reaction (PCR) is the most effective and accurate technique for detection of plant species as well as GMOs (Marmiroli et al., 2008). PCR analysis of food includes the following steps: sample preparation, DNA extraction, amplification of the target sequences by PCR and assessment of PCR products by agarose gel electrophoresis (Somma & Querci, 2006). The quality and quantity of the template DNA are critical factors for the successful PCR analysis. DNA integrity, purity and yield vary according to the material under examination and the DNA extraction method applied. Till date, numerous DNA extraction protocols have been developed; however, they have been rarely compared in a comprehensive manner (Zimmerman et al., 1998; Peano et al., 2004; Bernardo et al., 2007; Mafra et al., 2008). The comparative study of DNA extraction methods was focused on soybean and maize, because they are main GM crops; the raw materials as well as their derived food products were analysed. Peano et al. (2004) demonstrated that QIAamp DNA Stool mini kit and the Wizard method gave higher yield of genomic DNA than DNeasy Plant Minikit and Nucleo Spin Food kit for seeds and flours. However Mafra et al. (2008) evidenced that the extraction of soybean flour and simple products can be better

achieved by the kits NucleoSpin and GeneSpin than by the CTAB and Wizard Methods. Bernardo et al. (2007) verified that CTAB/PTB method and MasterPure DNA Purification kit (Epicentre) yielded the highest levels of DNA with a low level of quality, while High Pure GMO Sample Preparation kit (Roche Diagnostics GmbH, Mannheim, Germany) extracted the best amplifiable quality DNA for soybean and maize food products. Other authors found better performance of CTAB method than that of the Wizard method for the extraction of DNA from raw soybean and raw maize (Tung Nguyen et al., 2009). Cankar et al. (2006) revealed that the Wizard method exhibited high fluctuation in PCR efficiency. Recently Sagi et al. (2009) proved that Takara kit (Takara Bio Company, Madison, WI, USA) and enzymatic digestion method produced the best-quality DNA from rice cultivars.

The goal of this study was to determine the role the extraction methods play in DNA amplification in order to select the best protocol for several important food crops. For this purpose, three commonly used DNA extraction methods were compared for isolation and purification of genomic DNA from crop samples, such as soybean (Glycine max), maize (Zea mays), rice (Oryza sativa), wheat (Triticum aestivum), barley (Hordeum vulgare) and oats (Avena sativa). These plants are essential in human diet and domestic animal nutrition and are the most important crops for agriculture around the world. The current cereal transformation activities are focused on these crops. Moreover, soybean and maize are the most distributed genetically modified (GM) crops, while the area of transgenic rice has been growing increasingly for a few last years. Correspondingly, the reliable detection of these crops and/or their transgenic counterparts in food has a great importance for assessment of food quality and safety. The comparative study of genomic DNAs extracted by different protocols is in urgent need for development of accurate and efficient PCR-based

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