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Development of a Quantitative Real-time PCR-based Assay to Identify Adulteration in Commercial Turmeric Powder Products

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[Introduction]

Turmeric, or *Curcuma longa*, is commonly consumed in the South East Asian countries as a medical product and as food due to its therapeutic properties. However, with increasing demand for turmeric powder, adulterated turmeric powders mixed with other cheap starch powders, from corn or cassava. Here, we developed molecular markers using quantitative real-time PCR to identify adulteration in turmeric powder products. Chloroplast genes, such as *matK, atpF*, and *ycf2*, were used to develop species-specific primers for *C. longa* and *Zea mays.*

[Materials and Methods]

Turmeric (*Curcuma longa*) root tubers and corn (*Zea mays*) seeds were kindly provided by Gangwondo Agriculture Research and Extension Services (Chuncheon, Korea). All *C. longa* commercial products used for the analysis of food complexes were purchased from local markets and stored at room temperature. To generate a quantitative reference binary mixture model, binary mixtures containing different amounts (0. 1%, 1%, 10%, and 100%) of turmeric powders were prepared to a final mass of 2 g.

[Results and Discussion]

The efficiency of each primer set was within 90-110%. A linear correlation (R^2>0.98) were obtained in the 10-fold serial dilutions (10^7 to 10^3) of each recombinant plasmid and genomic DNAs (from 10 ng to 1 pg). We tested the sensitivity and specificity of the developed *C. longa* primer sets with binary mixtures (0.1-100% (w/w)) of *C. longa* dry powders containing each of three starch crops, including rice, corn, and wheat. All three *C. longa* primer sets with slopes ranging from -3.177 to -3.550 exhibited R^2> 0.99 and efficiency values of 91.29-106.43 % when used on mixed powders of *C. longa* and each starch crop, supporting the primer sets for verifying the presence of *C. longa* in mixtures. Subsequently, sensitivity of the three *Z. mays* primer sets was tested with binary mixtures of *Z. mays* and *C. longa* (0.1-100% (w/w)). Similarly, the three *Z. mays* primer sets with slope ranging from -3.220 to -3.437 exhibited R2> 0.99 and efficiency values of 95.41-104.44 % when used on mixed powders of *C. longa* and *Z. mays*, supporting the primer sets for verifying the presence of *Z. mays* in mixtures. The Ct values for the limit of detection (LOD) (0.1% target species in binary mixtures) were established to verify the presence of the target species. Ct values for the LOD ranged from 28 to 29 cycles for each primer set targeting *C. longa* and 28 to 29 cycles for those targeting *Z. mays*. We developed three chloroplast gene targeted primer sets for both *C. longa* and *Z. mays*. To assess the quantities of the target-species present, standard curves were constructed using recombinant plasmid DNA and binary DNA mixtures. Therefore, the developed qPCR assay could contribute to food safety and the protection of consumer's rights.

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