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Development of DNA-based Real-time PCR Analysis Method to Distinguish between *A. rugosa* and *P. cablin*, which can be used Interchangeably under the Name of “Kwakhyang,” and Validation of Applicability to Commercial Herbal Medicines

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[Introduction]

Agastache rugosa and *Pogostemon cablin* are commonly consumed as traditional herbs in East Asia. According to a report from the Ministry of Food and Drug Safety in 2022, a case was reported in which *A. rugosa* was used without distinguishing it from *P. cablin* under the name of “Kwakhyang” despite the difference in ingredients. Therefore, DNA-based species-specific PCR markers are needed to differentiate these two plants to ensure consumer rights.

[Materials and Methods]

DNA-based real-time analysis PCR assay was developed to detect *A. rugosa* and *P. cablin*, respectively. Species-specific primers were developed using chloroplast genes such as *matK*, *rbcL*, and *rpoB* and the ITS region of the nuclear DNA region. 15 commercial herbal medicines used in the study were purchased from local markets. Genomic DNA was extracted from commercial foods using a CTAB-based DNA extraction method.

[Results and Discussion]

The efficiency of each primer set ranged from 90-110%, and a linear correlation ($R^2 > 0.99$) was obtained between the crossover value and log DNA concentration. We determined the Ct values of 10 pg of the target species as the cutoff line and the Ct values of all non-target species amplified later than this cutoff line. Then, we evaluated the compatibility of the designed species-specific markers using 15 commercial herbal medicines. As a result of the test, all species-specific markers detected only the target species. Therefore, we expect that the real-time PCR analysis of this study will be usefully used to distinguish between *A. rugosa* and *P. cablin*.

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