Supporting Information to:

Application of Real-Time Scorpion PCR for Authentication and Quantification of the Traditional Chinese Medicinal Plant

*Drynaria fortunei*

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Detection Mixture of *Drynaria fortunei* and Adulterants

We mixed commercial *Drynaria fortunei*, *D. mollis*, and *D. sparsisora* to simulate the presence of *D. mollis* and *D. sparsisora* contaminant and isolated DNA from it. Species identification experiments were carried out with the scorpion of *D. fortunei*, *D. mollis*, *D. quercifolia*, *D. rigidula*, *D. sparsisora*, and *Pseudodrynaria coronans* (Fig. 5S). The results revealed that each of the scorpion probes is specific and is capable of definitely identifying the species. These results demonstrate that DNA identification and verification in herbal mixtures can be performed by multiple-scorpion real-time PCR.
**Fig. 1S** Aligned sequences of *trnL-trnF* from *Drynaria fortunei*, *D. sparsisora*, *D. rigidula*, *D. quercifolia*, *D. mollis*, and *Pseudodrynaria coronans*. Dashes are gaps required for alignment. The forward primer, reward primer, and the sequence that is complementary to the probe are indicated (arrows).
**Fig. 2S** Electrophoretic analysis of genomic DNA extracted from fresh leaf and commercially prepared crude drugs on 2% agarose gel. M: 100-bp DNA ladder (MW); 1: fresh leaf of *Drynaria fortunei*; 2: commercially prepared crude *Drynaria fortunei*.

**Fig. 3S** Amplification plot of the scorpion assay with *Drynaria fortunei* and *Pseudodrynaria coronans* mixtures on a fixed weight of total samples as templates. Amplifications with the *Drynaria fortunei*–specific scorpion probe.
Fig. 4S Matrix effects of *D. mollis*, *D. quercifolia*, *D. rigidula*, and *D. sparsisora* on *Drynaria fortunei* calibration curves. The PCR cycle number is plotted against the log percentage of the standard dilutions (10%, 25%, 50%, 75%, and 90%). The log percentage values are 1 (10%), 1.40 (25%), 1.70 (50%), 1.88 (75%), and 1.95 (90%), respectively.
Fig. 5S Real-time PCR assay of the mixed DNA samples of *Drynaria fortunei*, *D. mollis*, and *D. sparsisora*.