Hyper-sensitive Multiplex RNA Quantification System based on CE-SSCP

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Quantification of RNA provides the crucial information in various fields of biology, such as transcriptomics, pathogen detection. The major technology for RNA quantification is real-time PCR, which is known as the most accurate method of nucleic acids quantification. However, application of real-time PCR on RNA quantification is limited by single target per an analytical run because of reduced quantification power in multiplex detection and limitation on fluorescence dye.

Here, we report a novel multiplex RNA quantification method using capillary electrophoresis-single strand conformation polymorphism (CE-SSCP) coupled with asymmetric PCR using common primers. There are three major steps which are template modification, multiplex amplification, and simultaneous detection. Firstly, RNA targets are modified for multiplex amplification step. After the first step, all targets have the same end-sequences, and the sequences are the sites on which the common primers hybridize. Second step is multiplex amplification of targets using asymmetric PCR with the common primer pair. Finally, the amplicons are separated and quantified by CE-SSCP analysis.