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## **A method for detecting rash and fever illness-associated viruses using multiplex reverse transcription polymerase chain reaction.**

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### **Abstract**

In this study, a new multiplex RT-PCR method for detecting various viral genes in patients with rash and fever illnesses (RFIs) was constructed. New primer sets were designed for detection of herpes simplex viruses 1 and 2 (HSV1 and 2), and Epstein-Barr virus (EBV). The newly designed and previously reported primer sets were used to detect 13 types of RFI-associated viruses by multiplex RT-PCR assay systems. Moreover, to eliminate non-specific PCR products, a double-stranded specific DNase was used to digest double-stranded DNA derived from the templates in clinical specimens. RFI-associated viruses were detected in 77.0% of the patients (97/126 cases) by the presented method, multiple viruses being identified in 27.8% of the described cases (35/126 cases). Detected viruses and clinical diagnoses were compatible in 32.5% of the patients (41/126 cases). Sensitivity limits for these viruses were estimated to be  $10^1$  -  $10^3$  copies/assay. Furthermore, non-specific PCR products were eliminated by a double-stranded specific DNase with no influence on sensitivity. These results suggest that this method can detect various RFI-associated viruses in clinical specimens with high sensitivity and specificity.

**KEYWORDS:** Fever; multiplex RT-PCR; rash; virus

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