

**Quantitation of severe acute respiratory syndrome  
coronavirus genome by real-time polymerase chain  
reaction assay using minor groove binder DNA probe  
technology.**

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摘要

**Abstract**

The ability to rapidly recognize severe acute respiratory syndrome coronavirus (SARS-CoV) as a cause of infections is critical to quickly limiting further spread of the disease. A rapid, sensitive, and specific laboratory diagnostic test is needed to confirm outbreaks of SARS-CoV infection and distinguish it from other diseases that can cause similar clinical symptoms. An improved TaqMan technology using minor groove binder (MGB) probes was used to detect and quantify SARS-CoV in suspected patients. SARS-CoV primers and probe were designed based on the open reading frame 1b sequence, which encodes coronavirus replicase protein. A linear standard curve with  $R^2 > 0.99$  was obtained, and the threshold sensitivity was 10 genome equivalents per reaction. Interassay coefficients of variation were 1.73 to 2.72%, indicating good reproducibility of this method. A total of 228 specimens from 151 suspected patients were quantified by this method, 13 specimens from 6 patients were positive with viral load range from 362 to 36,240,000 genome equivalents/mL. In conclusion, the high sensitivity and reproducibility of the real-time polymerase chain reaction SARS-CoV RNA quantitation using MGB probe allowed the screening of large numbers of clinical samples.