

Bacteriology and Infectious Diseases

May 16-18, 2016 San Antonio, USA

Analytical validation of three alternative PCR master mixes for *Bordetella* species real-time PCR detection

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The Pertussis and Diphtheria Laboratory at the Centers for Disease Control and Prevention (CDC) developed a real-time PCR (rtPCR) assay for the detection of three *Bordetella* species. This assay is routinely used at the CDC, public health laboratories in the USA and internationally for pertussis diagnostics. It relies on one approved and validated PCR master mix, Applied Biosystems Taqman Gene Expression Master Mix (GE). In recent years PCR master mixes have been engineered to be resistant to PCR inhibitors, thus potentially providing more sensitive alternative master mixes to be used in this assay. Three commercially available alternative master mixes were identified: Quanta PerfeCTa SuperMix (QS), Quanta PerfeCTa ToughMix (QT) and Quanta PerfeCTa ToughMix with UNG (QTU). The performance of these master mixes was evaluated by comparing target-specific detection levels and consistency among three rtPCR instruments previously validated and currently used for routine pertussis diagnosis. The analytical sensitivity and specificity of these alternative PCR master mixes was assessed by the *Bordetella* species rtPCR multi-target assay. We report that QS produced Ct values comparable to GE, while QT and QTU produced lower Ct values (≥ 2 Ct value difference) and thus exhibited higher analytic sensitivity across all assays and instruments. Therefore, before selecting an alternative master mix or rtPCR platform, laboratories should validate their own diagnostic assay to ensure it is producing results with clinically relevant cut-offs, as those previously published.

Biography

Brunilis Burgos-Rivera has completed her PhD in Genetics from the University of Georgia in 2012. Currently, she is a Microbiologist at the Pertussis and Diphtheria Laboratory at the Centers for Disease Control and Prevention (CDC) working as a Contractor. She has served as the Laboratory Coordinator for the Latin American Pertussis Project in collaboration between CDC, Sabin Vaccine Institute, Pan American Health Organization and the Ministries of Health in select Latin American Countries to strengthen pertussis surveillance and diagnostics in the region. More recently, she has been selected as a Fellow to the CDC Laboratory Leadership Service, Class of 2016.

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