Project Title: Real-time PCR detection system for diarrheagenic *E. coli*.
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Abstract:

To compare the diarrheagenic *Escherichia coli* (DEC) identifications obtained between traditional O serotyping and modern virulence gene detection assays, we developed a multiplex real-time PCR assay by detecting six specific virulence genes for enteropathogenic *E. coli* (EPEC), enterohemorrhagic *E. coli* (EHEC), enterotoxigenic *E. coli* (ETEC), and enteroinvasive *E. coli* (EIEC). Among 386 clinical diarrheal stool samples, a total of 198 suspected DEC (sDEC) isolates were identified by the use of commercially available antisera. The most prevalent serogroups were O1 (23/198; 11.6%), O25 (15/198; 7.6%), and O44 (13/198; 6.6%). The specific virulence genes for the 198 sDEC isolates were analyzed by the multiplex real-time PCR assay. Sixteen (8.1%) of 198 isolates were confirmed to be true DEC strains, indicating that the serotypic markers did not correlate with the specific virulence genes. ETEC (62.5%) was the most prevalent, followed by EIEC (18.75%) and EPEC (18.75%). No EHEC strains were identified in the specimens. Four novel serotypes were found in the study: two in EPEC strains (O111:H9 and O63:H6) and two in EIEC strains (O63:H9 and O169:H9). In conclusion, the real-time PCR assay considerably reduces the high false-positive rate from the use of serotyping alone, and thus, it is suggested that serogrouping-based methods are inadequate for the identification of DEC isolates, although they are useful for the identification of a limited number of serogroups. In addition, ETEC, EPEC, and EIEC strains were present in 4.1% (16/386) of the diarrheal patients in northern Taiwan in 2006.

Keyword: real-time PCR, diarrheagenic *E. coli*, virulence gene, serotyping,