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Improved detection and quantitation of norovirus from water

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ABSTRACT

Norovirus is associated commonly with human sewage and is responsible for numerous cases of waterborne and foodborne gastroenteritis every year. Assays using reverse transcription-quantitative polymerase chain reaction (RT-qPCR) have been developed for norovirus, however, accurate detection and quantitation remain problematic owing to a lack of suitable positive controls. To improve enumeration of norovirus genomes from water, a synthetic norovirus genogroup II quantitation standard and competitive internal positive control were developed. The quantitation standard demonstrates identical amplification efficiency as wildtype norovirus and can be used as a viral surrogate in labs with restricted access to norovirus. The internal control quantifies sample inhibition, allowing for accurate quantitation of norovirus from complex environmental samples. Seawater samples spiked with sewage or bird guano were evaluated using the norovirus assay as part of a methods comparison study. Inhibition was detected in nine of 36 (25%) samples, two of which proved to be positive upon re-analysis. Results support the specificity of this assay for human-source (sewage) fecal contamination. Overall, use of this quantitation standard and internal control signify a great advance over traditional positive controls and suggest that molecular techniques for viral analysis could become standardized for routine water quality monitoring.