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Rapid QPCR-based assay for fecal *Bacteroides* spp. as a tool for assessing fecal contamination in recreational waters

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ABSTRACT

Concentrations of fecal indicator bacteria (FIB; e.g. *Escherichia coli*, and *Enterococcus* sp.) can only be used in limited ways for determining the source of fecal contamination in recreational waters because they cannot distinguish human from non-human fecal contamination. Several *Bacteroides* spp. have been suggested as potential alternative indicators. We have developed a rapid, culture-independent method for quantifying fecal *Bacteroides* spp. using quantitative PCR (QPCR) targeting the 16S rRNA gene. The assay specifically targets and quantifies the most common human *Bacteroides* spp. The details of the method are presented, including analyses of a wide range of fecal samples from different organisms. Specificity and performance of the QPCR assay were also tested via a laboratory experiment where human sewage and gull guano were inoculated into a range of environmental water samples. Concentrations of fecal *Bacteroides* spp., total *Enterococcus* sp., *Enterococcus faecium*, *Enterococcus faecalis*, and *Enterococcus casseliflavus* were measured using QPCR, and total *Enterococcus* sp. and *E. coli* were quantified by membrane filtration (MF). Samples spiked with gull guano were highly concentrated with total *Enterococcus* sp., *E. coli*, *E. faecalis*, and *E. casseliflavus*, demonstrating that these indicators are prominent in animal feces. On the other hand, fecal *Bacteroides* spp. concentrations were high in samples containing sewage and were relatively low in samples spiked with gull guano. Sensitivity and specificity results suggest that the rapid fecal *Bacteroides* spp. QPCR assay may be a useful tool to effectively predict the presence and concentration of human-specific fecal pollution