Monitoring bacterial faecal contamination in waters using multiplex real-time PCR assay for *Bacteroides* spp. and faecal enterococci

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Abstract

Monitoring of sanitary quality or faecal pollution in water is currently based on quantifying some bacterial indicators such as *Escherichia coli* and faecal enterococci. Using a multiplex real-time PCR assay for faecal enterococci and *Bacteroides* spp., the detection of faecal contamination in non-treated water can be done in a few hours, reducing the analysis time to 2 h.

The conventional method based on cultures was compared with a multiplex assay procedure for *Bacteroides* spp. and faecal enterococci with an internal inhibition control. Out of 74 water samples from different sources analyzed, using both procedures, 54 were true positives and 6 true negatives, 12 samples were real-time PCR positive and culture-negative whereas 2 were real-time PCR negative and culture-positive. In conclusion, 89.2% of the samples were found to be positive with real-time PCR and 75.7% with plate cultures.

Detection levels were much higher when using the multiplex real-time PCR assay, based on the higher number of positive samples in comparison with conventional microbiology. The feasibility of multiple reactions in the monitoring of faecal contamination has been demonstrated along with fast quantification of the faecal load. Such procedure can be performed in less than 3 h.

This work extends the use of multiplex real-time PCR for environmental analysis, demonstrating the feasibility of these procedures in monitoring faecal pollution of water samples.

Keywords: faecal contamination, multiplex real-time PCR, water monitoring, faecal enterococci, *Bacteroides* spp.