

Comparison of conventional culture and real-time quantitative PCR using SYBR Green for detection of *Legionella pneumophila* in water samples

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Abstract

The genus *Legionella* comprises more than 40 species and 64 serogroups with approximately half of those species associated with human diseases. *Legionella pneumophila* Serogroup 1 is the most common pathogenic species and is responsible for up to 80% of legionellosis cases in the world. *Legionella* levels in water are assessed routinely by culture on a selective medium, but its slow growth is a serious drawback, given that at least 10 days are required to obtain results. In an attempt to provide a simple screening method for *Legionella pneumophila* in water systems samples a real time PCR assay using SYBR Green was developed. A total of 50 samples from cooling towers and hot tap water systems were analysed by DNA amplification using 2 pairs of primers targeting the *mip* and *dot* genes. *Legionella pneumophila* Serogroup 1 (NCTC12821) was used as a reference strain and to evaluate real-time PCR performance. The assays were successful with both primer sets; good and similar amplification efficiencies were achieved. In addition, high sensitivity was obtained; the method proved to allow for the detection of fewer than 10 gene copies per reaction. Results of real-time PCR were compared to conventional analysis based on culture. Although no strong correlation was observed between both methods and consequently real-time PCR could not substitute for the reference method, it represents a powerful screening tool. The inexpensive, sensitive and rapid real-time PCR based in SYBR Green method is of interest in monitoring *Legionella pneumophila* contamination, especially in environmental samples, and should be economical for large-scale routine tests.

Keywords: *Legionella pneumophila*, real time PCR, SYBR Green