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Rapid identification of Acanthamoeba from contact lens case using loop-mediated isothermal amplification method

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Abstract

A method employing loop-mediated isothermal amplification (LAMP) of 18S ribosomal RNA gene was developed to detect Acanthamoeba in contact lens cases. A prevalence of 7% (10/150) was detected, with 100% sensitivity and 100% specificity when compared with the standard culture technique. Using visual inspection of turbidity a minimum of 10 pg of Acanthamoeba DNA could be detected, 10 times more sensitive than quantitative PCR employing two of the LAMP primers. The production of LAMP amplicons was confirmed by gel-electrophoresis and ethidium bromide staining. The LAMP procedure takes less than 2 h to perform and will be useful for incorporation into a point-of-care screening of suspected Acanthamoeba infection.

Author Keywords

Acanthamoeba keratitis; Contact lens; Loop-mediated isothermal amplification; Protozoa; Rapid identification; SYBR Green quantitative PCR

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