

Optimization and Evaluation of Rapid Methods for the Assessment of Waterborne *Escherichia coli* in Egypt

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Abstract

Three different methods were developed and evaluated for determination of water borne *E. coli* from two wastewater purification stations and one drinking water station at Cairo, Egypt. The most appropriate parameters used in the enzymatic method were found to be; using the substrate concentration (700 mgI⁻¹), temperature of the water bath shaker (44.5° C), pH (7.2), and the time of measurement (30 minutes). Moreover, there was no significant difference between the samples' enzymatic activity measured with or without addition of the inducer IPTG (isopropyl-β-D-1-thiogalactopyranoside). Multiplex PCR was used to detect *E. coli* that enables its differentiation from biochemically and phylogenetically related bacteria. The target genes have been increased to three genes: *uidA*, *lacZ* and *lacY*. Evaluation of these methods indicated a significant correlation between the microbial count and the enzymatic activity of β-D-galactosidase produced by *E. coli* when using chromogenic (Pearson r = 0.87) or fluorogenic substrate (Pearson r = 0.749). In conclusion, fluorometric method was suitable for detection of low contaminated samples, while colorimetric was suitable for highly contaminated sample, multiplex PCR based on three target genes was suitable for quiet identification of environmental *E. coli* either viable, VBNC (viable but nonculturable cells) or dead cells.

Keywords: β-D-galactosidase, *Escherichia coli*, evaluation, optimization, waterborne.

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Microbial Inactivation by Chlorine in the Presence and Absence of a Chlorine-Demanding Substrate and Its Effect on β -D-Galactosidase in Egypt

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Abstract

Chlorine is a powerful antimicrobial substance due to its potential oxidizing capacity. However, decrease of the actual chlorine level through the oxidation of organic matter contaminated water sources is observed. This study reveals that the germicidal effect (GE) of chlorine doses in aqueous contact media is significantly higher than that in organic matter containing contact media. The effect of chlorine on cultivability and β -D-Galactosidase (GALase) activity of E. coli isolates were compared and revealed that GALase activities were more resistant to chlorine than their cultivability. Therefore, the determination of enzymatic activity as a method for quantification of the waterborne pathogens is more suitable with the chlorinated water sources that may have stressed pathogens (can be called viable but are nonculturable (VBNC) bacteria) which do not have the ability to grow properly on the solid media.

Key words: β -D-Galactosidase, chlorine, Germicidal effect, viable but nonculturable cells.

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