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Analytical limits of four β-glucuronidase and β-galactosidase-based commercial culture methods used to detect *Escherichia coli* and total coliforms

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ABSTRACT

Colilert® (Colilert), Readycult® Coliforms 100 (Readycult), Chromocult® Coliform agar ES (Chromocult), and MI agar (MI) are β-galactosidase and β-glucuronidase-based commercial culture methods used to assess water quality. Their analytical performance, in terms of their respective ability to detect different strains of *Escherichia coli* and total coliforms, had never been systematically compared with pure cultures. Here, their ability to detect β-glucuronidase production from *E. coli* isolates was evaluated by using 74 *E. coli* strains of different geographic origins and serotypes encountered in fecal and environmental settings. Their ability to detect β-galactosidase production was studied by testing the 74 *E. coli* strains as well as 33 reference and environmental non-*E. coli* total coliform strains. Chromocult, MI, Readycult, and Colilert detected β-glucuronidase production from respectively 79.9, 79.9, 81.1, and 51.4% of the 74 *E. coli* strains tested. These 4 methods detected β-galactosidase production from respectively 85.1, 73.8, 84.1, and 84.1% of the total coliform strains tested. The results of the present study suggest that Colilert is the weakest method tested to detect β-glucuronidase production and MI the weakest to detect β-galactosidase production. Furthermore, the high level of false-negative results for *E. coli* recognition obtained by all four methods suggests that they may not be appropriate for identification of presumptive *E. coli* strains.

1. Introduction

The multiple-tube fermentation and membrane filter techniques are classical reference methods used for water quality monitoring (APHA et al., 1998) that have been associated with their own limitations. The multiple-tube fermentation method provides results only after 3 to 4 days and the interference by a high number of non-coliform bacteria have been shown to alter the efficiency of the analysis (Evans et al., 1981; Means and Olson, 1981; Seidler et al., 1981). For the membrane filter technique, the most widely used medium for drinking water analysis are m-Endo and mFC media in United States and Canada (APHA et al., 1998) and Tergitol-TTC medium in Europe (AFNOR, 1990). However, since these media lack specificity, coliform confirmation is required (APHA et al., 1998) which delays the results by 2 to 3 days. Also, the presence of a high number of background heterotrophic bacteria was shown to decrease coliform recovery (Burlingame et al., 1984; Clark, 1980). The inherent limitations of these two methods make them unable to provide, within hours, useful public health information.

To diminish background effects of heterotrophic bacteria and circumvent the need for a confirmation stage required by both multiple-tube fermentation and membrane filter techniques, methods based on the enzymatic properties of coliforms (β-galactosidase for total coliforms and β-glucuronidase enzymes for *Escherichia coli* detection) have been developed. These enzymes have been chosen because conventional coliform monitoring is based on detection of the presence of β-galactosidase and because the gene encoding the β-glucuronidase enzyme (*uidA*) was found to be specific (Brenner et al., 1972) and present in more than 97% of *E. coli* isolates (Lupo and Halpern, 1970; Martins et al., 1993).

Colilert® (Colilert, IDEXX Laboratories, Westbrook, ME, USA), Readycult® Coliforms 100 (Readycult; Merk KGaA, Darmstadt, Germany), Chromocult® Coliform agar ES (Chromocult; Merk KGaA, Darmstadt, Germany), and MI agar (MI; BD, Franklin Lakes, NJ, USA) are four commercial test methods based on the determination of β-galactosidase and β-glucuronidase enzyme activities which are used to detect, within 24 h, total coliforms and *E. coli* in water samples. These tests are easy to use, require no additional confirmatory step, and provide a more rapid estimate of indicators of bacteriological contamination of water as compared to classical techniques (Brenner

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Table 1
Ability of Colilert, Readycult Coliforms 100, Chromocult Coliform agar ES and MI agar test methods to detect *Escherichia coli* and *Shigella* sp. strains

Strains (origin)	No. reference	Serotype	Test methods			
			Colilert	Readycult Coliforms 100	Chromocult Coliform agar ES	MI agar
Escherichia coli (n=74)						
Escherichia coli (clinical)	ATCC 11775	O1:K1:H7	+	+	+	+
Escherichia coli (clinical)	ATCC 23511	O16:K1(L):NM	+	+	+	+
Escherichia coli (clinical)	ATCC 35401	O78:H11	+	+	+	+
Escherichia coli (clinical)	ATCC 43886	O25:K98:NM	−	+	+	+
Escherichia coli (clinical)	ATCC 43890	O157:H7	−	−	−	−
Escherichia coli (clinical)	ATCC 43894	O157:H7	−	−	−	−
Escherichia coli (clinical)	ATCC 43895	O157:H7	−	−	−	−
Escherichia coli (clinical)	ATCC 43896	O78:K80:H12	+	+	+	+
Escherichia coli (clinical)	CCRI-1191	N/A	+	+	+	+
Escherichia coli (clinical)	CCRI-1192	N/A	+	+	+	+
Escherichia coli (clinical)	CCRI-1193	N/A	+	+	+	+
Escherichia coli (clinical)	CCRI-1213	N/A	+	+	+	+
Escherichia coli (clinical)	CCRI-2099	N/A	+	+	+	−
Escherichia coli (clinical)	CCRI-2105	N/A	−	+	+	+
Escherichia coli (clinical)	CCRI-2106	N/A	−	+	+	+
Escherichia coli (clinical)	CCRI-2107	N/A	−	+	+	+
Escherichia coli (clinical)	CCRI-2108	N/A	−	+	+	+
Escherichia coli (clinical)	CCRI-2109	N/A	−	−	+	−
Escherichia coli (clinical)	CCRI-2166	N/A	−	−	−	−
Escherichia coli (clinical)	CCRI-2202	N/A	−	+	−	−
Escherichia coli (clinical)	CCRI-8825	N/A	−	+	+	+
Escherichia coli (clinical)	CCRI-8826	N/A	−	+	+	+
Escherichia coli (clinical)	CCRI-8831	O157:H7	−	−	−	−
Escherichia coli (clinical)	CCRI-8832	O157:H7	−	−	−	−
Escherichia coli (clinical)	CCRI-8833	O103:H2	−	+	+	+
Escherichia coli (clinical)	CCRI-8834	O103:H2	−	+	+	+
Escherichia coli (clinical)	CCRI-8835	O111:H-	−	+	+	+
Escherichia coli (clinical)	CCRI-8836	O111:H-	−	+	+	+
Escherichia coli (clinical)	CCRI-8837	O26:NM	−	−	−	−
Escherichia coli (clinical)	CCRI-8838	O26:NM	+	+	+	+
Escherichia coli (clinical)	CCRI-8839	O145:NM	−	−	+	+
Escherichia coli (clinical)	CCRI-8840	O145:NM	+	+	+	+
Escherichia coli (clinical)	CCRI-8852	N/A	−	−	−	+
Escherichia coli (clinical)	CCRI-9493	N/A	−	+	+	+
Escherichia coli (environmental)	CCRI-14813	N/A	+	+	+	+
Escherichia coli (environmental)	CCRI-14858	N/A	+	+	+	+
Escherichia coli (environmental)	CCRI-14859	N/A	+	+	+	+
Escherichia coli (environmental)	CCRI-14871	N/A	−	+	−	+
Escherichia coli (environmental)	CCRI-14881	N/A	+	+	+	+
Escherichia coli (environmental)	CCRI-16465	N/A	+	+	+	+
Escherichia coli (environmental)	CCRI-16485	N/A	+	+	+	+
Escherichia coli (environmental)	CCRI-16527	N/A	+	+	+	+
Escherichia coli (environmental)	CCRI-16528	N/A	+	+	+	+
Escherichia coli (environmental)	CCRI-16537	N/A	−	+	+	+
Escherichia coli (environmental)	CCRI-16539	N/A	+	+	+	+
Escherichia coli (environmental)	CCRI-16540	N/A	+	+	+	+
Escherichia coli (environmental)	CCRI-16579	N/A	+	+	+	+
Escherichia coli (environmental)	CCRI-16580	N/A	+	+	+	+
Escherichia coli (environmental)	CCRI-17006	N/A	+	+	+	+
Escherichia coli (environmental)	CCRI-17021	N/A	−	+	+	+
Escherichia coli (environmental)	CCRI-17027	N/A	−	+	+	+
Escherichia coli (environmental)	CCRI-17042	N/A	+	+	+	+
Escherichia coli (environmental)	CCRI-17045	N/A	+	+	+	+
Escherichia coli (environmental)	CCRI-17056	N/A	+	+	+	+
Escherichia coli (environmental)	CCRI-17063	N/A	+	+	+	+
Escherichia coli (environmental)	CCRI-17065	N/A	−	+	+	+
Escherichia coli (environmental)	CCRI-17097	N/A	+	+	+	+
Escherichia coli (environmental)	CCRI-17151	N/A	+	+	+	+
Escherichia coli (environmental)	CCRI-17158	N/A	+	+	+	+
Escherichia coli (environmental)	CCRI-17161	N/A	+	+	+	+
Escherichia coli (environmental)	CCRI-17172	N/A	−	+	+	+
Escherichia coli (environmental)	CCRI-17176	N/A	+	+	+	+
Escherichia coli (clinical)	LSPQ 2086	O8:H9	+	+	+	+
Escherichia coli (clinical)	LSPQ 2092	O18:NM	+	+	+	+
Escherichia coli (clinical)	LSPQ 2096	O26:NM	+	+	+	+
Escherichia coli (clinical)	LSPQ 2113	O111:NM	−	−	−	+
Escherichia coli (clinical)	LSPQ 2115	O128:H8	+	+	+	+
Escherichia coli (clinical)	LSPQ 2117	O113:H21	+	−	+	−
Escherichia coli (clinical)	LSPQ 2118	O117:H4	−	+	+	+
Escherichia coli (clinical)	LSPQ 2125	O128:NM	−	+	+	+
Escherichia coli (clinical)	LSPQ 2127	O157:H7	−	−	−	−
Escherichia coli (clinical)	LSPQ 3760	O157:H7	−	+	−	−
Escherichia coli (clinical)	LSPQ 3761	O157:H7	−	+	−	−

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