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# Comparison of Bacteriological Methods for Detecting and Enumerating Total Coliforms and *Escherichia coli* in Water

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## ABSTRACT

Bacteriological quality assurance of water using indicator bacteria, namely Total Coliforms (TC) and *Escherichia coli* (EC) is a routine practice around the world. The current study compared the performance of three alternative methods with the conventional Sri Lanka Standard (SLS)-Multiple Tube Fermentation (MTF) method, for the detection and enumeration of TC and EC in different tropical waters. Alternative methods included Colilert and m-ColiBlue24 methods and conventional SLS-Membrane Filtration (MF) method. Bottled water, well water, surface water and wastewater effluents (75 samples) were examined for TC and EC by the four methods, following ISO criteria. TC counts detected by all alternative methods were higher ($p \leq 0.05$) than with the conventional SLS-MTF method. Colilert ($p \leq 0.05$) and m-ColiBlue24 ($p \leq 0.1$) detected significantly higher EC counts compared to SLS-MTF. Simple Linear Model showed several folds higher TC counts by Colilert (3.85), m-ColiBlue24 (1.75) and SLS-MF (1.55) and EC counts by m-ColiBlue24 (1.83), Colilert (2.93) and SLS-MF (1.35) than the SLS-MTF. Confirmational rates were higher in all alternative methods, than SLS-MTF. ISO performance criteria (sensitivity, specificity and efficiency) were superior in all alternative methods compared to SLS-MTF. Enzymatic methods showed superior features in simultaneous detection of TC and EC (e.g., absence of background growth or atypical colonies; easy preparations and interpretations; lesser time and labour requirement etc.). In conclusion, the current study revealed that Colilert, m-ColiBlue24 and SLS-MF methods could be recommended as alternative methods for analyzing drinking and surface water samples in a tropical country like Sri Lanka.

**Key words:** Coliforms, colilert, *Escherichia coli*, m-ColiBlue24, MF, MTF

## INTRODUCTION

The bacteriological quality of water is monitored by the detection of indicator organisms, namely Total Coliforms (TC) and *Escherichia coli* (EC). The Multiple Tube Fermentation (MTF) which gives

bacterial counts in Most Probable Number (MPN) values and the Membrane Filtration (MF) methods have been widely used around the world for the detection and enumeration of these indicator bacteria (APHA, AWWA and AEF, 2000; Rompre *et al*., 2002). These two detection and enumeration methods of TC and EC are based on the conventional definition of these organisms as capable of fermenting lactose at 37 and 44°C, respectively (Fricker and Fricker, 1996). The presumptive detection of these organisms for lactose fermentation is done by observing growth, acid and gas production (in MTF method) and by acid production (in MF method). Confirmation of particular organisms is conducted by subsequent tests done for production of acid and gas from lactose at 37°C (for TC) and simultaneous detection of indole reaction together with acid and gas formation by lactose fermentation at 44.5°C (for EC) (SLS 614, 1983; APHA, AWWA and AEF, 2000).

However, the production of gas and acid from lactose fermentation has long been irrelevant since all coliforms and certain EC strains do not ferment lactose (Fricker and Fricker, 1996; Edberg *et al*., 2000). Both lactose-positive and lactose-negative biotypes have been isolated, irrespective of their origin (Leclerc *et al*., 2001). Furthermore, a significant proportion of the coliforms are reported to be an aerogenic, without ability to produce gas from lactose fermentation (Fricker and Fricker, 1996; Edberg *et al*., 2000) and approximately 10% of coliforms do not ferment lactose (Fricker *et al*., 1994).

Therefore, other detection methods with superior performance criteria such as higher sensitivities, specificities, efficiencies and lower false positive and false negative ratios have been introduced. Enzymatic methods based on the detection of enzymatic activities of the indicator bacteria using chromogenic and fluorogenic media have gain popularity among the alternative detection methods (Pitkanen *et al*., 2007). These methods detect the presence of **β**-D galactosidase and **β**-D glucuronidase enzyme activities in TC and EC, respectively (Rompre *et al*., 2002).

Though Sri Lanka is a developing country, it has a relatively high HDI (Human Development Index). With the aim of providing safe drinking water regular bacteriological monitoring of water is conducted. The Sri Lanka Standards Institute (SLSI) has established bacteriological water quality standards to safeguard the drinking water quality for consumption (SLS 614, 1983). The standards have recommended two standard methods, MTF method and the MF method, based on lactose fermentation ability of the coliform group for the analysis of TC and EC in water. Therefore, all the water quality monitoring institutes in Sri Lanka strictly adhere to the SLSI standards and follow the recommended MTF or MF procedures for detecting bacteriological quality of water. However, these methods are time and labour consuming. Therefore, analysis of large numbers of samples at the same time is a tedious task. Also the dearth of expert technical staff and the shorter shelf lives of reagents in hot and humid climate are major constraints in developing quality assurance of bacteriological testing in tropical countries like Sri Lanka. Therefore, adoption of more efficient, rapid and easy methods is essential. Further, the accuracy and sensitivity of the currently available SLS-MTF method in detecting and enumeration of TC and EC has not been adequately investigated or compared with the existing SLS-MF method or with other novel technologies in Sri Lanka. Therefore, the aim of this study was to investigate the performance of currently available conventional SLS-MTF method, compared with the existing SLS-MF method and other two alternative enzymatic methods for detecting and enumerating TC and EC in different tropical water types in Sri Lanka. Method comparison was conducted by following the ISO criteria for establishing equivalence between microbiological methods (ISO 17994: 2004).

## MATERIALS AND METHODS

**Bacteriological examination of water:** Water samples (sample number = 75) were collected from different sources (bottled, well, surface water and wastewater effluent), from different geographical areas of the country, as recommended by the International Standard (ISO 17994, 2004) and analyzed (in duplicate) using four methods, following the guidelines described by the Standard Methods for the Examination of Water and Wastewater (APHA, AWWA and AEF, 2000) and Sri Lanka Standard 614: Part 2: 1983 (SLS 614, 1983). Sampling was conducted from July 2008 to June 2009.

**Sri Lanka Standards (SLS 614, 1983) MTF method:** Used as the reference method MacConkey broth (Oxoid, UK) was used for detecting TC presumptively, (incubated at 36±1°C for 24 to 48 h) by formation of acid and gas. Positive presumptive TCs were confirmed in Brilliant Green Lactose Bile broth (BGLB) (Oxoid, UK) incubated at 36±1°C for 24 to 48±2 h. Confirmation of EC was done by inoculating parallel tubes of BGLB and Peptone water (Oxoid, UK) incubated at 44.5°C for 24 to 48 h. Presence of EC was confirmed by detecting gas formation in BGLB and immediate appearance of a red colour ring after addition of 0.2-0.3 mL of Kovac's reagent (Oxoid, UK) on to Peptone medium.

**Sri Lanka Standards (SLS 614, 1983) MF method:** Appropriate volumes and dilutions of water samples were filtered through the membrane filtration apparatus (Pyrex, Germany) using sterilized membrane filters with 0.45 μm pore sizes (Sartorius, Germany). Membrane filters were aseptically placed on pre sterilized absorbent pads (Sartorius, Germany), saturated with 3 mL of M-endo medium (Himedia, India) and 3 mL of M-FC medium (Himedia, India) and were incubated at 36±1 and at 44.5°C (for 24 h), for the detection of TCs and EC, respectively.

**Colilert method (IDEXX, USA):** MPN technique was followed as described in the Standard Methods for the Examination of Water and Wastewater (APHA, AWWA and AEF, 2000), by using Colilert medium (IDEXX, USA) with appropriate dilutions. Appropriate weights of Colilert powder (IDEXX) were mixed with appropriate volumes of each dilution of the test samples. Sterilized distilled water was added to each tube to obtain the final volume and mixed well (colourless after mixing) and were incubated at 36±0.5°C for 24 h or less. A yellow colour after incubation was considered as a positive TC test and florescence under UV illumination (366 nm) was considered as EC positive.

**m-ColiBlue24 method:** Appropriate dilutions and volumes of different water samples were filtered though membrane filters and placed on absorbent pads saturated with 2.5 mL of m-ColiBlue 24 broth (Hach, USA) and incubated at 37°C for 24 h. Red colour TC colonies and blue colour EC colonies were counted and recorded as cfu/100 mL.

**Confirmation of bacteria:** TCs and EC were confirmed by following the ISO 9308-1 standard (ISO 1908-1, 2000), using pure isolated colonies obtained by sub-culturing on Tryptic Soy Agar (TSA) (Oxoid, UK) plates.

**Reference cultures:** *Escherichia coli* ATCC 19522 and *Pseudomonas fluorescens* (laboratory isolate) were used as positive reference cultures in each test.

**ISO Performance criteria (ISO/TR 13843):** Performance of methods was assessed by following the ISO criteria for Guidance on validation of microbiological methods (ISO/TR 13843). The performance characteristics (sensitivity, specificity, false positive rate, false negative rate and efficiency) of four methods were analyzed for TC and EC.

**Comparison of methods according to ISO criteria (ISO 17994, 2004):** In each comparison, counts (confirmed) from the same sample obtained on the alternative method were paired to counts (confirmed) obtained on the reference medium (SLS-MTF). After natural logarithmic transformation of the paired count data, the relative difference ($X_i$) between compared methods in each sample were calculated using the equation $X_i = \ln (a_i) - \ln (b_i) \times 100$ % according to ISO 17994 (2004), where $a_i$ and $b_i$ are paired counts from different methods. The mean relative difference ($\bar{x}$) was counted by the equation $\bar{x} = \Sigma X_i/n$. Thus, it is the sum of relative differences divided by the number of samples (n).

**Statistics:** SAS System for Windows V8 software (SAS Institute Inc., SAS Online Doc®, Version 8, Cary, NC) was used for statistical analysis of data. The differences were evaluated statistically significant in cases where p-values were = 0.05 or = 0.10.

## RESULTS

**Bacteriological counts obtained by different methods:** The four bacteriological analytical methods detected different counts (cfu/100 mL) of TC and EC for the same water sample. Total values of TC and EC present in different water sources during the experimental time period were obtained by summing up the bacteriological counts detected in each sampling, for different sources. The total values obtained were plotted against methods, for different source water types (Fig. 1).

Mean bacteriological counts obtained by the alternative methods for all water sources (grouped together) were compared with that of the SLS-MTF reference method by using Least Squares Mean Separation test (Table 1). The experimental design used was Complete Randomized Block Design (CRBD), while the source water types used in the study were blocked in analysis. Both MF and MPN results were transformed in to logarithmic values for statistical analysis.

**Simple linear model for analyzing the relationships between different methods:** The relationship between different alternative methods with the SLS-MTF reference method was analyzed by using the Simple Linear Model test. Results of the linear relationships between SLS-MTF reference method with the other alternative methods for the detection and enumeration of TCs and EC are shown in Table 2.

**Confirmation rates of bacteria by different methods:** Confirmation rates for TC and EC were obtained by dividing the number of confirmed tests by the number of tubes or colonies used in confirmation tests multiplied by 100. Results are depicted in Table 3.

**ISO performance criteria obtained by different methods:** In detecting TC, m-ColiBlue24 and SLS-MF showed the highest sensitivities (1.0 and 0.9, respectively). The specificity of Colilert method (0.47) was higher than that of the SLS-MF (0.18). False positive rate was highest in SLS-MTF (0.29). False positive rate was lowest in Colilert (0.15), when detecting TC. Further, the false negative rates were zero in both enzymatic methods (Colilert and the m-ColiBlue24) since the two

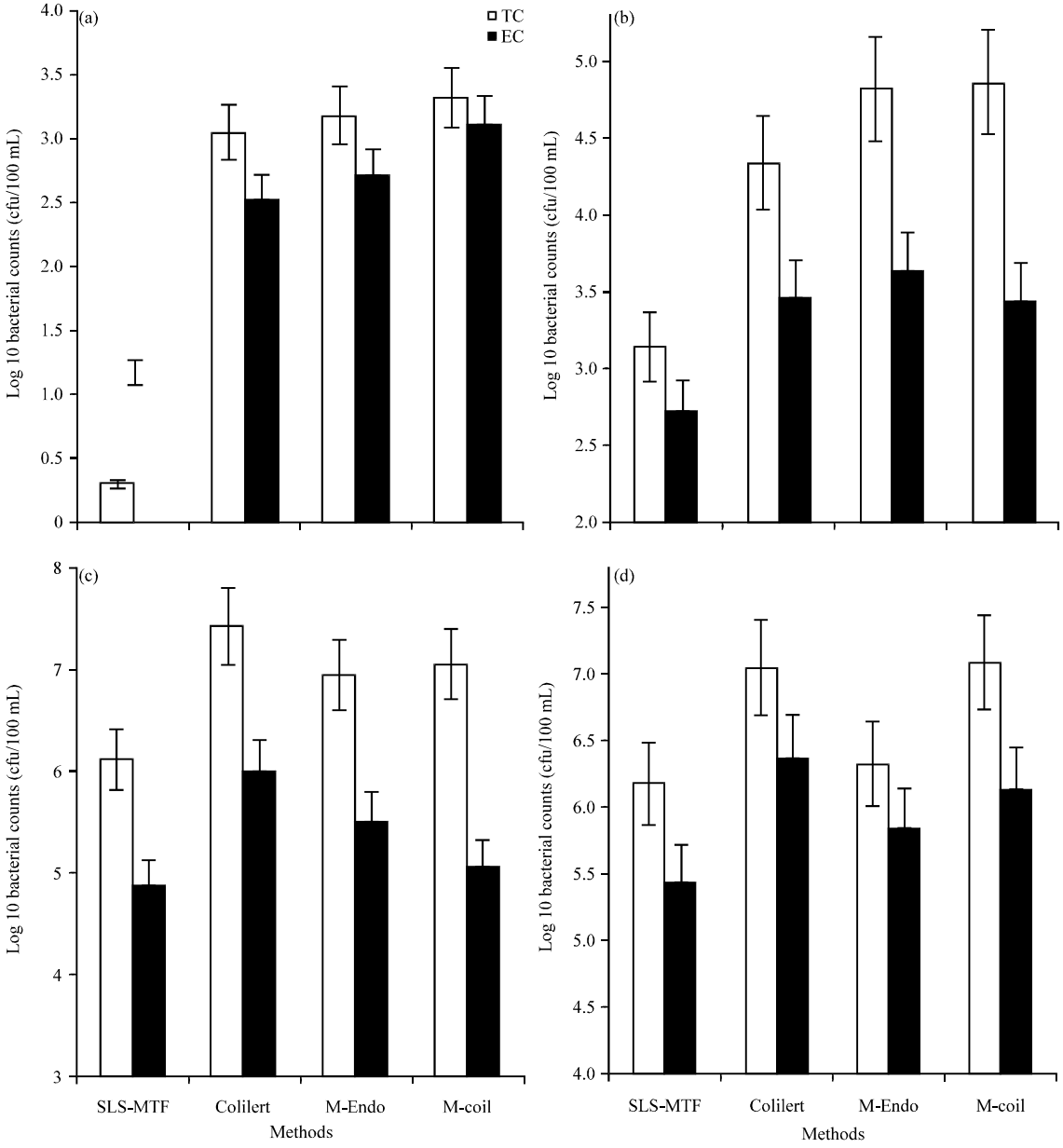


Fig. 1(a-d): Total values of TC and EC counts (log $_{10}$ values) in different water sources obtained by four methods (from July 2008 to June 2009) (a) Bottled water, (b) Well water, (c) Surface water and (d) wastewater effluent, TC-Total colliforms, EC-*Escherichia coli*

ColiBlue24 and SLS-MF methods, respectively. Furthermore, the false negative rates were zero in all alternative methods. The highest efficiency among the methods was shown with Colilert method (0.39), compared to other methods. It was 0.36 and 0.28 in SLS-MF and m-ColiBlue24 methods, respectively, while the minimum rate was obtained with the reference SLS-MTF method (0.13).

**Comparison of methods according to ISO criteria (ISO 17994, 2004):** Results obtained with method comparison using paired counts are shown in Table 4. Evaluation of the performance of

Table 1: Results of the least squares mean separation test to compare alternative methods with SLS-MTF reference method

| | p-value for comparison with SLS-MTF | |
|---|---|---|
| Comparison | TC | EC |
| SLS-MTF/Colilert | 0.0021* | 0.0280* |
| SLS-MTF/M-endo/M-FC | <.0001* | 0.1581 |
| SLS-MTF/m-ColiBlue24 | <.0001* | 0.0823** |

*p≤0.05, **p≤0.1

Table 2: Results of the simple linear relationships for detecting TC and EC

| | Lines for TC counts | | Lines for EC counts | |
|---|---|---|---|---|
| Method tested to SLS-MTF method | Slope of the line y=k x | $R^2$ | Slope of the line y=k x | $R^2$ |
| Colilert | 3.853 | 0.916 | 1.837 | 0.986 |
| M-FC or M-endo | 1.553 | 0.995 | 1.356 | 0.902 |
| m-ColiBlue24 | 1.753 | 0.984 | 2.936 | 0.984 |

The equations y=k x, where y is TC or EC counts obtained by alternative method. x = TC or EC counts obtained by the SLS-MTF method. R squired = coefficient of determination

Table 3: Confirmation test results for detecting total coliforms and *E. coli*

| | TC | | EC | |
|---|---|---|---|---|
| Method | Type of observation | Confirmation rate (%) | Type of observation | Confirmation rate (%) |
| SLS | Gas formation/ (and acid formation) | 71 | Positive indole reaction | 37.5 |
| Colilert | Yellow colouration | 78.2 | Fluorescence | 66.6 |
| M-endo/M-FC | Typical red metallic | 75.1 | Typical blue | 50 |
| | sheen/pink cream | 60 | Cream | 0 |
| m-ColiBlue | Typical red | 72.1 | Typical blue | 50 |

Table 4: Method comparison with SLS-MTF method for enumeration of TC and EC

| | Method comparison | | | | | |
|---|---|---|---|---|---|---|
| | SLS-MTF/Colilert | | SLS-MTF/M-endo/M-FC | | SLS-MTF/m-ColiBlue24 | |
| Parameter | TC | EC | TC | EC | TC | EC |
| $\bar{x}$ | 2.9 | 17.3 | 6.3 | 27.7 | 0.7 | 19.8 |
| $x_L$ | -25.8 | -8.8 | -25.8 | -2.5 | -22.2 | -6.3 |
| $x_H$ | 31.8 | 43.5 | 31.7 | 57.9 | 23.6 | 46 |
| Evaluation | Inconclu | Inconclu | Inconclu | Inconclu | Inconclu | Inconclu |

$\bar{x}$ = Mean Relative difference; $x_L$ Lower limit; $x_H$ Upper limit

alternative methods, in relation to the reference SLS-MTF method for the enumeration of two methods did not show any presumptively negative test result. Furthermore, the highest efficiency (0.81) among all methods was shown by Colilert method. Next was m-ColiBlue24 (0.73); SLS-MF showed a 0.64 efficiency.

For EC enumeration, all alternative methods obtained a value of 1.0 for sensitivity. Specificity was 0.26 in Colilert method, where as it was only 0.1 in SLS-MF. The highest false positive rate was obtained by SLS-MTF method (0.86). Further, the minimum false positive rate was given by

Colilert method as in the case of detecting TC. False positive rates were 0.72 and 0.68 in m-bacterial types, was done by using the equations given in ISO standard (ISO 17994, 2004). A value of 10% as the maximum acceptable deviation from zero (D) was used in this study and the evaluations were done two-sided. Results are shown in Table 4.

## DISCUSSION

**Method comparison:** All three alternative methods detected higher TC and EC counts compared to the SLS-MTF method (Fig. 1). Statistical analysis showed that all the alternative methods were able to detect significantly higher TC counts at $p \leq 0.05$ (Table 1). In detecting EC, only the two enzymatic methods detected significantly higher counts (Colilert: $p \leq 0.05$, m-ColiBlue24: $p \leq 0.1$), while the SLS-MF counts were not significantly higher. Linear regression analysis also showed that the all three alternative methods detected several fold higher TC counts (Table 3). Similar observations were also reported by Covert *et al.* (1989), Noble *et al.* (2004) and Schets *et al.* (2002).

There can be several possible reasons for the above observations. One possibility might be due to the presence of an aerogenic (without producing gas) lactose fermenting coliforms in most of the samples analyzed (Eckner, 1998). Therefore, the SLS-MTF method might be unable to detect the coliforms that cannot produce gas in fermenting lactose (Leclerc *et al.*, 2001). On the other hand, Colilert can detect β-D galactosidase enzyme of TCs (Edberg *et al.*, 1988; Pitkanen *et al.*, 2007). According to Schets *et al.* (2002), coliform bacteria have both (for β-galactosidepermease enzyme) and *lacZ* (for β-galactosidase enzyme) genes. Certain coliforms have only gene responsible for lactose fermentation, while, the other coliforms do not have *lacY* but do have *lacZ*. Those bacteria do not ferment lactose but can use Colilert substrate ONPG (o-nitrophenyl β-D galactopyranoside). The third reason for higher recoveries of TCs and EC with the enzymatic methods in the present study might be due to the presence of stressed cells (McFeters *et al.*, 1997).

In the current study, the M-endo medium which is not an enzymatic method, was also able to detect significantly higher ($p<0.05$) TC counts compared to SLS-MTF method. Among the membrane filter media, M-endo has been reported to show superior sensitivity and selectivity, by producing equivalent results to MTF (APHA), even with chlorine-stressed coliforms (Tobin *et al.*, 1980). The higher results of M-endo in this study could be attributed to the efficiency of detecting the acid reaction by M-endo medium, due to its higher specificity and sensitivity than the MacConkey medium (SLS-MTF) in detecting the lactose fermentation reaction.

m-ColiBlue24 method also detected significantly higher ($p<0.05$) TC counts than the SLS-MTF method in this study. Similar observations have been demonstrated by Grant (1997), in comparison of M-endo with m-ColiBlue24, for detecting TCs.

The positive test in SLS-MTF method was mostly based on detecting gas formation, since, the acid reaction was not prominent in most of the tubes. Therefore, absence of gas formation in tubes is usually considered as negative presumptive tests causing false negative results in conventional SLS-MTF method.

EC enumeration test results also showed that, the SLS-MTF method detected the lowest mean counts compared to other methods. However, only the two enzymatic methods showed significantly higher counts of EC. Although the counts detected by M-FC were comparatively higher than the SLS-MTF, the difference was not significant. Similar observation has been also reported by Noble *et al.* (2004) and Griffith *et al.* (2006) in detecting EC in coastal waters.

In detecting EC, conventional SLS-MTF method uses the ability of EC to hydrolyze tryptophan to indole in the presence of pyruvic acid and ammonia. However, the low detection levels of EC in

SLS-MTF method, might be due to the missed EC, by the false negative results (gas negative), shown in the presumptive tests. Another reason might be due to the low performance of the peptone water broth.

Colilert method detected significantly higher ($p<0.05$) EC counts in this study. In most of the reported studies, EC concentrations detected by Colilert method showed no significant differences with the conventional MTF methods, in contrast to the results obtained in the current study (e.g., APHA-MTF method: Edberg *et al.* (1989) for surface water; Noble *et al.* (2004) for beach water samples; Griffith *et al.* (2006) for ambient water samples and the Swedish-MTF method: Eckner, (1998) for environmental waters). However, the difference observed in the current study, might be due to the high rate of detection of EC by the enzymatic activity in the tropical waters. Significantly higher EC counts detected by m-ColiBlue24 method in this study could be due to the detection of the enzyme β–glucuronidase present in EC cells, by 5-bromo-4-chloro-3-indolyl-b-D-glucuronic acid present in m-ColiBlue24 medium (Grant, 1997). This observation also proves the explanation by Pitkanen *et al.* (2007). The results of the current study indicated that the use of m-ColiBlue24 is an alternative approach that could provide better and more rapid information for the assessment of microbial quality of water. It could simultaneously detect TC and EC from a water sample within 24 h and hence, will provide utilities a quick measure of whether a sample has been subjected to fecal contamination.

Further, the findings of this study are in agreement with other studies which have compared the classical standard method procedures with enzymatic commercial kits. MI agar is one commercial preparation which has also shown higher performances compared to the conventional MTF methods (Brenner *et al.*, 1996). A study by Nikaeen *et al.* (2009) has indicated that the enzymatic LMX broth assay has recovered 1.4 and 1.18 times as many TC and EC, respectively, as the MTF method.

**Performance of methods:** The highest TC confirmation rate (78.2%) shown by the Colilert method in this study, is comparatively lower with other previous studies with different water types; 93.7% in subtropical fresh water samples (Chao *et al.*, 2004), 100% (Fricker and Fricker, 1996) and 90% (Pitkanen *et al.*, 2007) in temperate waters. Further, the false positive rate of Colilert was 15% in this study. These observations might be due to the detection of non coliform bacteria causing false positive results (Covert *et al.*, 1989). However, in the current study, the colourless tubes did not contain any TC bacteria making the false negative rate zero.

Being a specific and sensitive medium, M-endo might have specifically detected more TC than the lactose based medium MacConkey. However, the atypical cream colour colonies formed on M-endo plates were also confirmed as TC (60%). Therefore, even though M-endo was sensitive (sensitivity = 0.9) in detecting TC, its specificity was 0.18 in this study. Present results showed a false positive rate of 24% which is similar presented by Grant (1997) but higher than Evans *et al.* (1981). In present study, the efficiency of the M-endo medium in detecting TC was comparatively low with the Colilert method. In m-ColiBlue24 method there was no atypical colony formation observed in this study, compared to the finding of Grant (1997) who observed 1.6% atypical colonies. Due to the absence of atypical colonies on m-ColiBlue24 plates, the sensitivity was 100% in detecting TCs in present study. However, it showed 28% false positive TCs, as similarly reported by Grant (1997). However, in the current study, there were no Gram positive bacteria detected by m-ColiBlue24 method in this study. It shows that the m-ColiBlue24 is specific only to gram negative bacteria which is a positive feature, observed in this method. The efficiency of m-ColiBlue24 in TCs

in this study was 0.73 which was comparatively lower with the Colilert method but higher than the other lactose based methods. These false positive results shown in both M-endo and m-ColiBlue24 might be due to the detection of non-coliform species as described above for Colilert method. Highest EC confirmation rates shown by Colilert might be due to the specificity of the enzymatic activity in detecting EC. The lowest confirmation rate and the highest false positive rate (86%) given by the SLS-MTF method might be due to the low specificity of the medium to detect EC. Further, surprisingly highest proportion (13.6%) of Gram-positive bacteria was also detected by this method in this study. Therefore, the overall non-coliform bacterial percentage detected by SLS-MTF was 63.6% which was the highest among the four methods compared.

**Method comparison by ISO criteria:** The current study was the pioneering study which involved the incorporation of ISO criteria for comparison of bacteriological methods in Sri Lanka and probably the pioneering study in South East Asia. Bacteriological comparisons, following the ISO criteria and using fabricated samples created primarily from laboratory strains of bacteria seeded into clean matrices and then, by Pitkanen *et al.* (2007), using natural non-disinfected water samples.

Observations in this study, revealed that all the comparisons resulted in 'inconclusive' evaluations for both TC and EC, although more than hundred confirmation tests were conducted as prescribed by the ISO. This was due to several facts such as problems arose with the natural environmental water samples, loss of cultures due to poor growth and poor isolations due to culture contamination which finally resulted in obtaining a lower number of paired count data. It became the major constraint in this comparison study causing inconclusive evaluations. These results suggested a requirement for increasing the number of samples by all four methods, as described by ISO criteria. Since there were no previous records on similar work done by using SLS-MTF method, results obtained in this study could not be compared. However, one such study done previously, by Pitkanen *et al.* (2007) by following the ISO criteria, has reported inconclusive evaluations even with comparatively higher sample numbers.

## CONCLUSIONS

In conclusion, the bacteriological methods examined in the current study performed differently, independent on the water source or the sampling location. Different strengths and weaknesses of the methods in detecting TCs and EC were identified. Qualitative and quantitative analyses of the current study proved that the existing conventional SLS-MTF method has many limitations in detecting and enumeration of TC and EC in water. In contrast, other alternative methods showed superior performance in detecting both bacterial types accurately. These alternative methods were more sensitive, specific and efficient than the SLS-MTF. Further, based on the contamination level and water sources, appropriate methods could be selected depending on cost effectiveness and the other performance criteria such as sensitivity, specificity, efficiency and other common features like simplicity, rapidity, user-friendliness, etc. of the method. In comparison, the SLS-MF also proved many advantages compared to the SLS-MTF method in the current study. Therefore, for routine bacteriological quality analysis, SLS-MF method which is currently in use, might be a better alternative than the conventional SLS-MTF method. Furthermore, the current study revealed that m-ColiBlue24 and Colilert methods, with their superior performance could be recommended as alternative methods for analyzing drinking and surface water, respectively, when the cost is not the limiting factor.

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