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Towards in line monitoring of coliforms in potable and recreational water

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Coliforms remain to be a valuable indicator of fecal contamination of surface water. Communities counting on surface water as a source of drinking or potable water depend on the regular monitoring of the source water for the presence of potential pathogenic microorganisms. The same applies to recreational water as it also becomes a public health concern. There have been several developments in sensor technologies and testing methods during the past decade including DNA based tests but the classical enzyme based methods are robust and remain to be one of the best methods. Current testing in some communities still involves the regular manual collection of water samples for analysis of coliform presence in the laboratory. The availability of a small footprint benchtop unit that can do this testing can potentially pave the way to develop autonomous systems to perform and report real time on these tests or using in line system integration.

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Detection of pathogens by polymerase chain reaction: Comparison with bacteriological water testing kit and standard methods for water quality monitoring

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A defined substrate media as bacteriological water testing kit (BWTK) has been developed to detect the presence/absence of total coliforms, fecal coliforms, *Aeromonas hydrophila*, *Yersinia enterocolitica* and *Listeria* spp., from drinking water. A total of 10,000 drinking water samples from municipal corporation supply, submersible pumps and hand pumps were analyzed by both methods, BIS 10500:1991 and BWTK, at 5% level of significance no difference was observed. Microbiological and physicochemical analysis of 4000 drinking water samples from gastroenteritis prone areas of three different utilities. In contaminated drinking water samples, *Aeromonashydrophila* 78.94%, *Yersinia enterocolitica* 69.61% and *Listeria* spp., 52.63% were observed. All the isolates were further characterized biochemically and at molecular level by using species specific primers. On the basis of nucleotide homology 16S rRNA gene and phylogenetic analysis, the isolate of *Aeromonas hydrophila* (GP1), *Yersinia enterocolitica* (GP2) and *Listeria* spp. (GP3) were submitted to NCBI, USA with an accession number GU596499, GU596500 and JF798637 respectively. It was found that occurrence of emerging pathogens are independent of occurrence of fecal coliforms, E. coli in water. All the isolates from water showed MAR indices >0.2. Mineral analysis revealed that concentration of Pb, Cu, Fe, Cr, K, Na, Co and Ca in drinking water samples, analyzed were much below the permissible limits, Ni detected in 57.14% of samples. Growth kinetic of *E. coli*, *Aeromonas hydrophila*, Yersinia enterocoitica and *Listeria* spp., in microcosm of water drawn from tap, millipore filter, mineral water, tap water with biofilm and under psychrophilic conditions (4 °C) revealed *E. coli* as short lived whereas *Y. enterocolitica* adapted better to all the environmental conditions. A combined culture, BWTK and multiplex polymerase chain reaction (PCR) a molecular tool, as water testing kit has been designed for simultaneous detection of *Yersinia enterocolitica*, *Aeromonas hydrophila* and *E. coli* in drinking water. The optimized conditions for multiplex PCR water testing kit are: Annealing temperature 57 °C, Mg2+ concentration 3.0 mM, concentration of primer pairs of *E. coli*, *Aeromonas hydrophila* and *Yersinia enterocolitica* are 0.166 µM, 0.333 µM and 0.416 µM, respectively. In vivo study revealed that these emerging pathogens produced histopathological changes in liver, lung, heart, intestine and kidney. A biochemical kit for specific detection of emerging pathogens has been developed. Highest MIC of Sodium hypochlorite (4%) required by *Yersinia enterocolitica* (20 ppm) followed by *Listeria* spp., (10 ppm), *Aeromonas hydrophila* (6 ppm), *E. coli* (6 ppm) and contact time of 30 minutes. TEM analysis of water samples revealed presence of virus like particle of varying size 18-1726.72 nm.

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