

## Molecular Detection of E. coli in Treated Wastewater and Well Water Samples Collected from Al Riyadh Governorate, Saudi Arabia

**Authors :** Hanouf A. S. Al Nuwaysir, Nadine Moubayed, Abir Ben Bacha, Islem Abid

**Abstract :** Consumption of waste water continues to cause significant problems for human health in both developed and developing countries. Many regulations have been implied by different world authorities controlling water quality for the presence of coliforms used as standard indicators of water quality deterioration and historically leading health protection concept. In this study, the European directive for the detection of Escherichia coli, ISO 9308-1, was applied to examine and monitor coliforms in water samples collected from Wadi Hanifa and neighboring wells, Riyadh governorate, kingdom of Saudi Arabia, which is used for irrigation and industrial purposes. Samples were taken from different locations for 8 months consecutively, chlorine concentration ranging from 0.1- 0.4 mg/l, was determined using the DPD FREE CHLORINE HACH kit. Water samples were then analyzed following the ISO protocol which relies on the membrane filtration technique (0.45µm, pore size membrane filter) and a chromogenic medium TTC, a lactose based medium used for the detection and enumeration of total coliforms and E.coli. Data showed that the number of bacterial isolates ranged from 60 to 300 colonies/100ml for well and surface water samples respectively; where higher numbers were attributed to the surface samples. Organisms which apparently ferment lactose on TTC agar plates, appearing as orange colonies, were selected and additionally cultured on EMB and MacConkey agar for a further differentiation among E.coli and coliform bacteria. Two additional biochemical tests (Cytochrome oxidase and indole from tryptophan) were also investigated to detect and differentiate the presence of E.coli from other coliforms, E. coli was identified in an average of 5 to 7 colonies among 25 selected colonies. On the other hand, a more rapid, specific and sensitive analytical molecular detection namely single colony PCR was also performed targeting hha gene to sensitively detect E.coli, giving more accurate and time consuming identification of colonies considered presumptively as E.coli. Comparative methodologies, such as ultrafiltration and direct DNA extraction from membrane filters (MoBio, Germany) were also applied; however, results were not as accurate as the membrane filtration, making it a technique of choice for the detection and enumeration of water coliforms, followed by sufficiently specific enzymatic confirmatory stage.

**Keywords :** coliform, cytochrome oxidase, hha primer, membrane filtration, single colony PCR

**Conference Title :** ICSR 2020 : International Conference on Scientific Research and Development

**Conference Location :** Chicago, United States

**Conference Dates :** December 12-13, 2020