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Identification of Virulent Gene Clusters (*Eae-A* and *Stx2*) in *Escherichia coli* O157 Isolated from the Bolgoda River

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Abstract

The Bolgoda River, the largest freshwater reservoir in Sri Lanka's Western Province, flows through the Colombo and Kalutara districts and supports diverse domestic and industrial uses. Rapid urbanisation and industrialisation have contributed to significant pollution, creating conditions that favour the survival and dissemination of pathogenic microorganisms, including *Escherichia coli*. Among pathogenic strains, *E. coli* O157 is of particular concern due to its production of Shiga toxins (*stx1*, *stx2*) and intimin (*eae-A*), which can cause severe gastrointestinal disorders such as haemolytic uremic syndrome and haemorrhagic colitis. The present study aimed to detect the virulence genes *stx2* and *eae-A* in *E. coli* O157 isolated from 16 sites along the Bolgoda River, to assess water quality and public health risk. Water samples were processed to isolate *E. coli* on Eosin Methylene Blue (EMB) agar, followed by DNA extraction using the freeze-thaw method. PCR amplification with *stx2* and *eae-A* specific primers, followed by gel electrophoresis, was used to identify target virulence genes. Out of the forty DNA samples tested, three isolates were positive for *stx2*, all originating from the site near the Karadiyana dumpsite, suggesting leachate contamination as a likely source. In contrast, no isolates tested positive for *eae-A*. These results indicate the presence of pathogenic *E. coli* O157 in the Bolgoda River, highlighting the potential public health risk and underscoring the need for continuous microbial monitoring and improved wastewater and landfill management to safeguard water quality in this critical freshwater system.

Keywords: *Bolgoda river, E. coli O157, Virulent genes, stx2, eae-A*