Open Reading Frame Marker-Based Capacitive DNA Sensor for Ultrasensitive Detection of Escherichia coli O157:H7 in Potable Water

Authors: Rehan Deshmukh, Sunil Bhand, Utpal Roy

Abstract: We report the label-free electrochemical detection of Escherichia coli O157:H7 (ATCC 43895) in potable water using a DNA probe as a sensing molecule targeting the open reading frame marker. Indium tin oxide (ITO) surface was modified with organosilane and, glutaraldehyde was applied as a linker to fabricate the DNA sensor chip. Non-Faradic electrochemical impedance spectroscopy (EIS) behavior was investigated at each step of sensor fabrication using cyclic voltammetry, impedance, phase, relative permittivity, capacitance, and admittance. Atomic force microscopy (AFM) and scanning electron microscopy (SEM) revealed significant changes in surface topographies of DNA sensor chip fabrication. The decrease in the percentage of pinholes from 2.05 (Bare ITO) to 1.46 (after DNA hybridization) suggested the capacitive behavior of the DNA sensor chip. The results of non-Faradic EIS studies of DNA sensor chip showed a systematic declining trend of the capacitance as well as the relative permittivity upon DNA hybridization. DNA sensor chip exhibited linearity in 0.5 to 25 pg/10mL for E. coli O157:H7 (ATCC 43895). The limit of detection (LOD) at 95% confidence estimated by logistic regression was 0.1 pg DNA/10mL of E. coli O157:H7 (equivalent to 13.67 CFU/10mL) with a p-value of 0.0237. Moreover, the fabricated DNA sensor chip used for detection of E. coli O157:H7 showed no significant cross-reactivity with closely and distantly related bacteria such as Escherichia coli MTCC 3221, Escherichia coli O78:H11 MTCC 723 and Bacillus subtilis MTCC 736. Consequently, the results obtained in our study demonstrated the possible application of developed DNA sensor chips for E. coli O157:H7 ATCC 43895 in real water samples as well.

Keywords: capacitance, DNA sensor, Escherichia coli O157:H7, open reading frame marker

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