A microfluidic filter biochip-based chemiluminescence biosensing method for detection of Escherichia coli O157:H7

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Abstract: A chemiluminescence biosensing method combined with a microfluidic filter biochip was investigated and evaluated for rapid and sensitive detection of Escherichia coli O157:H7, A microfluidic filter biochip was designed based on stepped filter configuration to concentrate and form a single layer of immunomagnetic microbeads inside a reaction microchamber. The filter biochip was assembled by thermally bonding two glass chips (microchamber and microchannel chips) together. The microchamber chip with one inlet and a reaction microchamber was 1 mm x 7 mm x 11.5 mu;m, and the microchannel chip with three outlets was 1 mm x 1 mm x 2.5 μm. Carboxyl-modified magnetic microbeads (8.27 μm diameter) covalently coupled with anti-E. coli O157:H7 antibodies were used for the separation of target bacteria from the background. The food sample containing E. coli O157:H7 was mixed with immunomagnetic microbeads and horseradish peroxidase-labeled anti-E. coli O157:H7 antibodies to form sandwich complexes. A syringe pump was used to inject the sandwich complexes into the filter biochip, and then luminol was added to generate a chemiluminescence signal, which was collected, measured, and recorded in real time through a fiber optic light guide connected to a photon detector coupled to a PC with a data acquisition unit. The results indicated that this filter biochip-based chemiluminescence biosensing method could detect as few as 71 cells of E. coli O157:H7 inside the reaction microchamber of 12 nL volume by single-batch sampling without pre-enrichment. The volume of sample used for testing was 100 μL. A multi-batch sampling technique was used to increase the capture efficiency of the immunomagnetic microbeads for detecting low numbers of E. coli O157:H7, which reduced the detection limit to 34 cells of E. coli O157:H7. The total detection time was 90 min.

Author Keywords: Bacterial detection; Chemiluminescence; E. coli O157:H7; Microbeads; Microchannel; Microfluidic; Sampling technique

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