Analytical Microsystems for the Detection of 4 Dengue Virus Serotypes

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Abstract

A serotype-specific bioanalytical microsystem was developed for the rapid detection of Dengue virus (serotypes 1, 2, 3, and 4) in blood samples. The microsystem is an integration of an electrochemical biosensor with the isothermal Nucleic Acid Sequence-Based Amplification (NASBA) technique that is designed to amplify small amounts of RNA at 41 oC. The principle of the biosensor is based onan earlier dipstick-type optical biosensor developed in our lab for Dengue virus detection. It is based on DNA/RNA hybridization and liposome amplification. Two specific probes are designed to hybridize with the Dengue virus RNAs. One probe is immobilizedto ferri/ferrihexacyanide encapsulating liposomes, the other is immobilized on magnetic beads. The probes immobilized on the magnetic beads are serotype specific, while the probes on the liposomes can hybridize to all four Dengue virus serotypes. The electrochemical microbiosensor consists of 6 channels that are fabricated with the photoresist SU-8 or PDMS, two micromixer sections within the channel structures, interdigitated ultramicroelectrode arrays (IDUA) as transducer, a magnet and several input and output wells. Liposomes, magnetic beads and viral RNA are mixed in the micromixers, and hybridization complexes are captured on the magnet. The amount of liposomes on the magnet correlates thus directly to the concentration of viral RNA. A detergent is added to the channels, which lyses the liposomes caught on the magnet. The liposome detergent solution is mixed thoroughly in a second micromixer before being detected and quantified on the IDUAs. Viral RNA concentrations in the low fmol range can be detected with this highly sensitive bioanalytical microsystem.