Electrochemical Detection of DNA Mutations on a PNA-Modified Electrode Utilizing a Single-Stranded DNA Specific Endonuclease

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A novel electrochemical method to identify DNA mutations has been developed. First, target samples hybridize with the PNA(peptide nucleic acid) capture probe, resulting in current decrease in cyclic voltammogram(CV) by [Fe(CN)6]3-/4-. This decrease is a consequence of charge repulsion by the negatively charged target DNA. Single-stranded DNA specific endonuclease, nuclease S1, is then applied to the PNA/DNA duplex. When the hybridized PNA/DNA duplex contains a mismatched site, the target DNA is hydrolyzed by the nuclease S1 and removed from the electrode surface. This event leads to recovery of the current to the same level that existed before target hybridization. In contrast, the decreased current from matched PNA/DNA duplex is not recovered because the target DNA is not acted upon by the nuclease. By using this strategy, we were able to detect various mutation types, including single base insertion, deletion and substitution, in the BRCA 1 gene. The approach developed in this study exhibits reliable discriminating power between wild type and mutant samples and, as a result, represents a new strategy for the electrochemical diagnosis of human genetic mutations.