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Exonuclease III–assisted cascade signal amplification strategy for label-free and ultrasensitive electrochemical detection of nucleic acids

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In this work, a simple, signal-on and label-free electrochemical biosensor for ultrasensitive DNA detection is reported on the basis of an autocatalytic and exonuclease III (Exo III)-assisted cascade signal amplification strategy. In the presence of target DNA (T-DNA), the hybridization between the 3'-protruding DNA fragment of hairpin DNA probe (HP1) and T-DNA triggered the Exo III cleavage process, accompanied by the releasing of T-DNA and autonomous generation of new DNA fragment which was used for the successive hybridization with the another hairpin DNA (HP2) on the electrode. After the Exo III cleavage process, numerous quadruplex-forming oligomers which caged in HP2 were liberated on the electrode surface and folded into G-quadruplex-hemin complexes with the help of K⁺ and hemin to give a remarkable electrochemical response. As a result, a low detection limit of 4.83 fM with an excellent selectivity toward T-DNA was achieved. The developed electrochemical biosensor should be further extended for the detection of a wide spectrum of analytes and has great potential for the development of ultrasensitive biosensing platform for early diagnosis in gene-related diseases.

Keywords: Electrochemistry; DNA biosensor; Exonuclease III; signal amplification strategy

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