

Method of sensitive electrochemical detection by Using Isothermal Probe Amplification Based on EXPAR

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We describe a method of sensitive electrochemical detection by using isothermal probe amplification based on EXPAR (exponential amplification reaction). Due to low abundance of target nucleic acid, many isothermal amplification methods have been developed for improving sensitivity. EXPAR can be considered as suitable for short target. And electrochemical methods have received attention due to its advantages of cost, speed, simplicity, sensitivity and requires no optical instrument. In this case, a capture probe (CP) was immobilized on the gold electrode. And it can hybridize with methylene blue (MB)-labeled signaling probe (SP). Then, when presence of target, SP release from CP-SP complex. SP will make complex with trigger probe (TP) blocked by blocker probe (BP). Nicking enzyme recognizes the specific sequence on SP-TP complex and nicks the double stranded complex followed by the polymerization from nicked site, which can induce exponential amplification of probe that can act as primer on CP-SP complex. Repeat the EXPAR on the CP-SP complex, SP released to solution lead to MB derived peak current decrease. Finally, we interested in development of electrochemical sensor for POCT.