

# Screening of broad-spectrum aptamer and development of electrochemical aptasensor for simultaneous detection of penicillin antibiotics in milk

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**Abstract:** Penicillin antibiotics (PENs) play an important role in killing pathogenic bacteria. However, the residues of various penicillin antibiotics in milk gradually accumulate in the human body with the increase of milk intake, which causes direct harm to the human body. Aptamers can be used as recognition element of sensors. It is great significance to use broad-spectrum aptamers for simultaneous detection of PENs. In this study, we reported the screening and identification of DNA aptamers for PENs. The aptamers were screened by graphene oxide-systematic evolution of ligands by exponential enrichment (GO-SELEX). The broad-spectrum aptamers with high affinity and specificity were successfully obtained after 13 rounds of screening. The affinity and specificity of candidate aptamers were analyzed by a GO fluorescence competition method. Further sequence analysis revealed that a truncated 47 nt aptamer (P-11-1) had a higher affinity than the original 79 nt aptamer. The truncated aptamer P-11-1 was used as a recognition element, and an electrochemical aptasensor was prepared using gold nanoparticles (AuNPs) combined with ferroferric oxide-multi walled carbon nanotube (Fe<sub>3</sub>O<sub>4</sub>-MWCNTs) complex. The results showed that the developed aptasensor achieved the simultaneous detection of PENs in milk samples across a concentration range of 2 nM–10,000 nM, achieving a limit of detection of 0.667 nM. This methodology provided a simple and sensitive new thinking for antibiotic multi-residue detection.

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