Electronic Supplementary Material

A Competitive Electrochemical Aptamer-Based Method for Aflatoxin B1 Detection with Signal-off Response

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Pretreatment of the gold electrode

Firstly, a gold electrode (2 mm in diameter) was polished to a mirror-like surface by using 0.05 μm alumina slurries on a microcloth, and then rinsed by ultrasonication in water and ethanol, respectively. Next, the electrode was electrochemically cleaned in 0.5 M H_2SO_4 aqueous solution by an oxidation step (2 V for 5 s) and then a reduction step (-0.35 V for 10 s). After that, cyclic voltammetry (CV) in the range of -0.35 V to 1.55 V was conducted (20 scans at a scan rate of 4 V/s, followed by 10 scans at a scan rate of 0.1 V/s). Then, a second electrochemical cleaning was performed in 0.1 M H_2SO_4 aqueous solution containing 0.01 M potassium chloride, to further clean the gold electrode surface. In details, a set of CV scans were conducted in four different potential ranges: (a) 0.2 to 0.75 V; (b) 0.2 to 1.0 V; (c) 0.2 to 1.25 V; (d) 0.2 to 1.5 V. Ten segments for each potential range, at a scan rate of 0.1 V/s. Finally, the gold electrode was rinsed with deionized water and blow-dried by nitrogen, and it was ready for DNA modification.

Table S1 CV characterization of different modified electrodes

Electrodes	Oxidation peak		Reduction peak	
	Potential (V vs. Ag/AgCl)	i_p value (μA)	Potential (V vs. Ag/AgCl)	i_p value (μA)
Au	0.27	23.03	0.16	-21.21
cDNA/Au	0.35	6.77	0.05	-4.15
MCH/cDNA/Au	0.38	5.81	-0.02	-2.59

Table S2 Comparison of some aptamer-based assays for AFB1

Detection method	Strategy	Dynamic range	LOD	Ref.
Colorimetry	Relying on assembly of an aptamer and two split DNAzyme halves	0.3 nM-31.2 μM	0.3 nM	[23]
Fluorescence	Fluorescence resonance energy transfer (FRET) quenching of fluorescence of CdTe quantum dots by AuNPs	10-400 nM	3.4 nM	[24]
Fluorescence	Using a fluorophore-labeled aptamer and its cDNA sequence that is labeled with a quencher	16-312 nM	5 nM	[25]
Fluorescence	A dipstick assay based on a competitive reaction of the biotin-modified aptamer between AFB1 and Cy5-modified DNA probes	1-31 nM	1 nM	[26]
Fluorescence	Based on quenching of the fluorescence of CdTe quantum dots by graphene oxide (GO)	1.6 nM-160 μM	1.4 nM	[27]
Electrochemistry	Using cystamine-poly (amidoamine) dendrimers layers as platform for immobilization of aptamer	0.4-10 nM	0.4 nM	[18]
Electrochemistry	Aptamers modified screen-printed carbon electrode (SPCE)	0.4-50 nM	0.4 nM	[28]
Electrochemistry	Using MB-tagged aptamer on a SPCE modified with functionalized graphene oxide	0.2-19 nM	0.16 nM	[29]
Electrochemistry	Electrochemical enzyme-linked oligonucleotide array	0.32-32 nM	0.28 nM	[30]
Electrochemistry	Based on competition between MB-labeled aptamer and complementary DNA modified on gold electrode	2-500 nM	2 nM	This work
Surface plasmon resonance	Aptamer surface plasmon resonance sensor	0.4-200 nM	0.4 nM	[22]

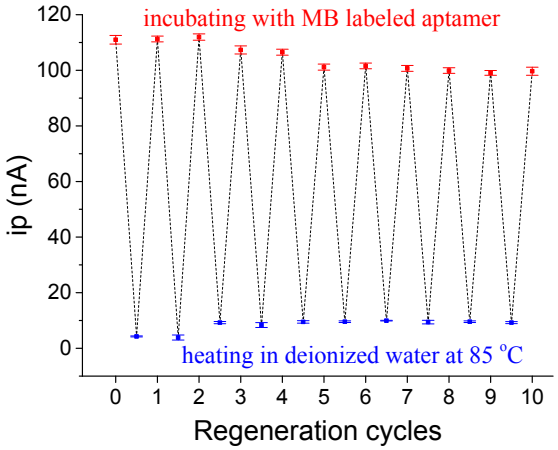

Fig. S1. Effects of regeneration cycles on i_p values of this electrochemical sensor. Experimental conditions: 50 nM MB-labeled aptamer, 20 mM $MgCl_2$ in binding buffer, and incubation for 5 min at 4 °C. The MB-labeled aptamer/MCH/cDNA/Au electrode was regenerated to MCH/cDNA/Au electrode by incubation in deionized water at 85°C for 1 min.

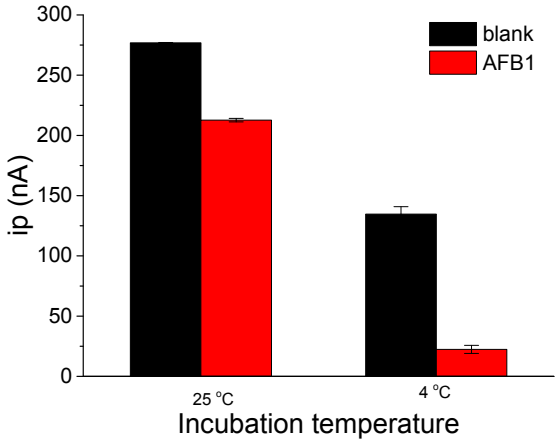

Fig. S2 Effect of incubation temperature on i_p value in the absence of or in the presence of AFB1 (200 nM). Experimental condition: 50 nM MB-labeled aptamer, binding buffer containing 20 mM $MgCl_2$, and incubation for 5 min.

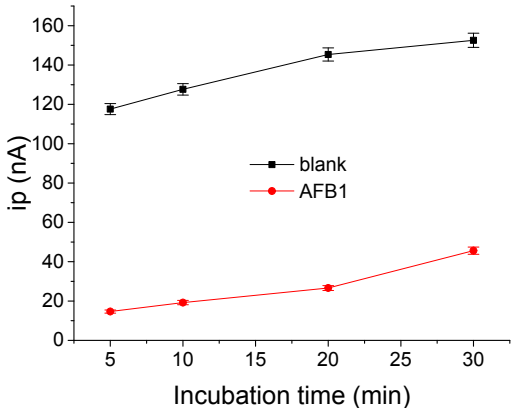

Fig. S3 Effect of incubation time on i_p values in the absence of or in the presence of AFB1 (200 nM). Experimental condition: 50 nM MB-labeled aptamer, binding buffer containing 20 mM $MgCl_2$, and incubation at 4 °C.

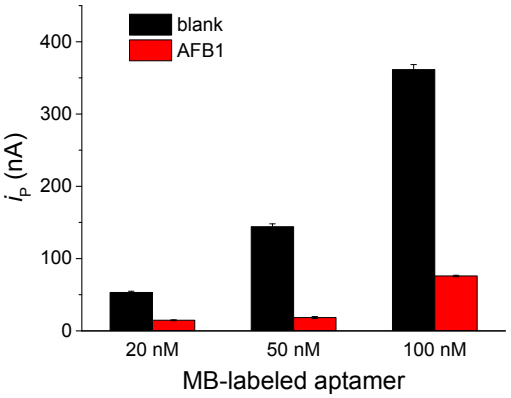

Fig. S4 Effect of concentration of MB-labeled aptamer in sample solution on i_p values in the absence of or in the presence of AFB1 (200 nM). Binding buffer containing 20 mM $MgCl_2$ and incubation for 5 min at 4 °C were applied.

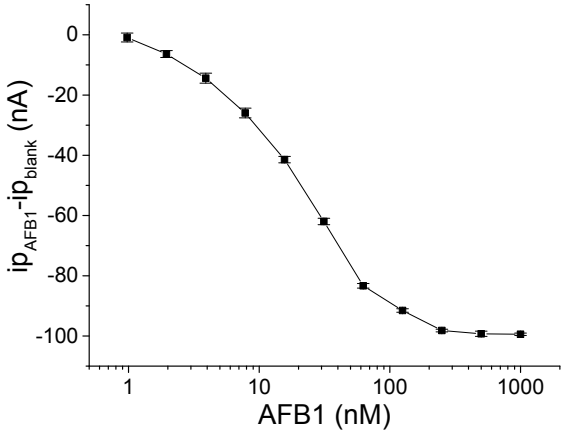

Fig. S5. Detection of AFB1 spiked in 50-fold diluted white grape wine.