Electronic Supplementary Information (ESI) for

A Label-free, Naked-Eye Detection of Aflatoxin B1 and Zearalenone Using "Naked" DNAzyme

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Table S1 The sequences are listed in the electronic supplementary information

Name	Sequence (5'-3')
S.DNAzyme	GGGCGGGTAGGGCGGG
1XAV	TGAGGGTGGGTAGGGTGGGTAA
1XAV-2	GGGTGGGTAGGGTGGG
A1-1	GGCACGTGTTGTCTCTCTGTGTCTCGTGCCTTTTTTAGACAACACGT GCAGGG
D1-3	ACAAGTGCGGGTAGGGCGGGAAGCACGTGTTGT

Table S2 The limit of detection (LOD) of AFB1 using S.DNAzyme in adlay, platyclade semen, tangerine peel and milk samples

	Adlay	Platycladi semen	Tangerine peel	Milk
LOD (μM)	1.4	0.78	1.17	2.3

Table S3 The limit of detection (LOD) of ZEN using S.DNAzyme in fritillaria cirrhosis bulbous, angelica sinensis, amygdalin, and rice samples

	Fritillariae cirrhosae bulbus	Angelica sinensis	Amygdalin	Rice
LOD (μM)	1.29	1.32	1.1	1.43

Table S4 Comparison of S.DNAzyme-based assay to other assays for colorimetric AFB1 or ZEN detection

Sensing element	Analytes	Label free	Enzyme free	Antibody free	LOD (μM)	Visual detection	Turnaround time (min)	Ref
S.DNAzyme	AFs (AFB1, AFB2, AFG1, AFM1) and ZEN	Yes	Yes	Yes	0.18[a] 0.29[b]	Yes	15	This work
Hairpin DNA probe assisted by exonuclease III	AFB1	Yes	No	Yes	1×10^{-6}	Yes	> 60	[1]
Polystyrene dyed particles conjugated with aptamer	AFB1	No	Yes	Yes	0.0146	Yes	35	[2]
Magnetic nanoparticle conjugated with monoclonal antibody	ZEN	No	No	No	1.3×10^{-4}	No	45	[3]
AuNPs conjugated with antibody	ZEN	No	Yes	No	NA	Yes	15	[4]
AuNPs conjugated with antibody	AFB1, OTA, ZEN	No	Yes	No	8.0×10^{-4}[a] 3.1×10^{-4}[b]	Yes	20	[5]
Different-colored AuNPs labeled with various monoclonal antibodies	ZEN, FB1, OTA, AFB1	No	Yes	No	1.9×10^{-4}[a] 2.2×10^{-3}[b]	Yes	NA	[6]
Aptamer and AuNPs	ZEN	Yes	Yes	Yes	0.031	Yes	15	[7]

OTA: ochratoxin, AFB1: fumonisin B1, [a]The LOD value of AFB1, [b]The LOD value of ZEN.

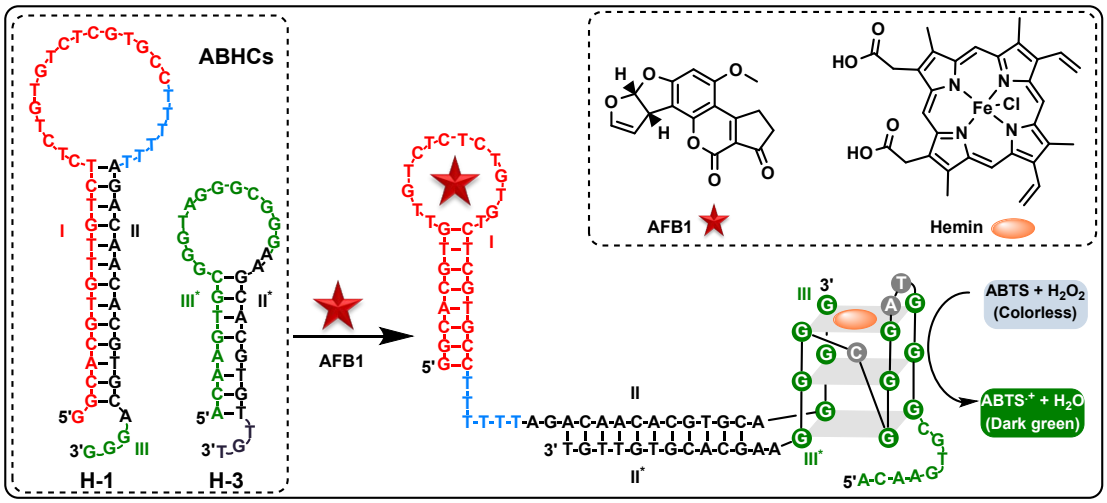

Fig. S1 Design and prediction of two aptamer-based hairpin constructs (ABHCs) for AFB1 detection.

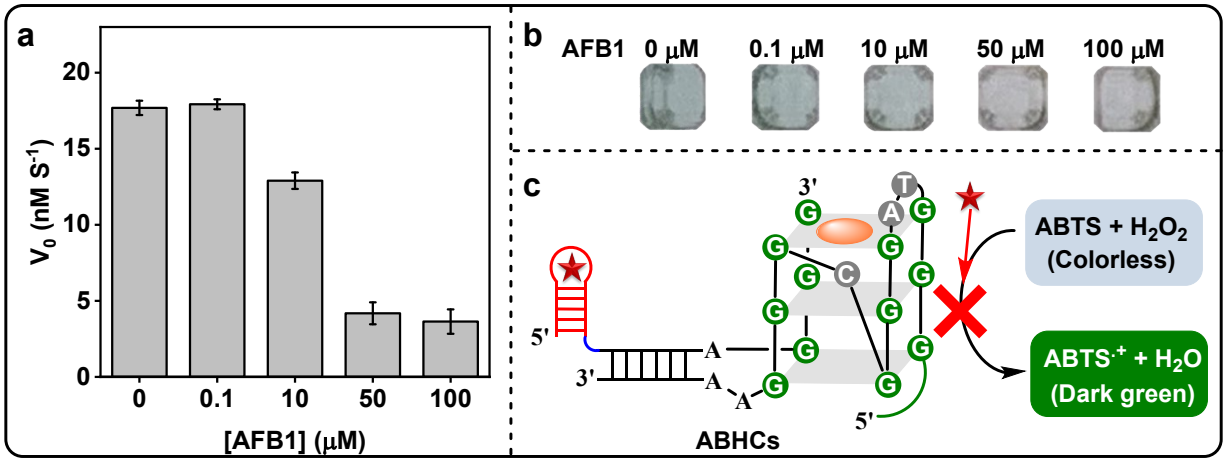

Fig. S2 The catalytic activity of ABHCs, 500 nM H-1 and H-3, in presence of different concentration of AFB1. (a) Reaction rates of ABHCs in terms of nanomolar $ABTS^{\bullet+}$ produced per second. (b) Photographs of samples containing 0−100 μM AFB1 after 15 min of reaction. (c) Scheme for the predicted reaction between ABHCs and AFB1.

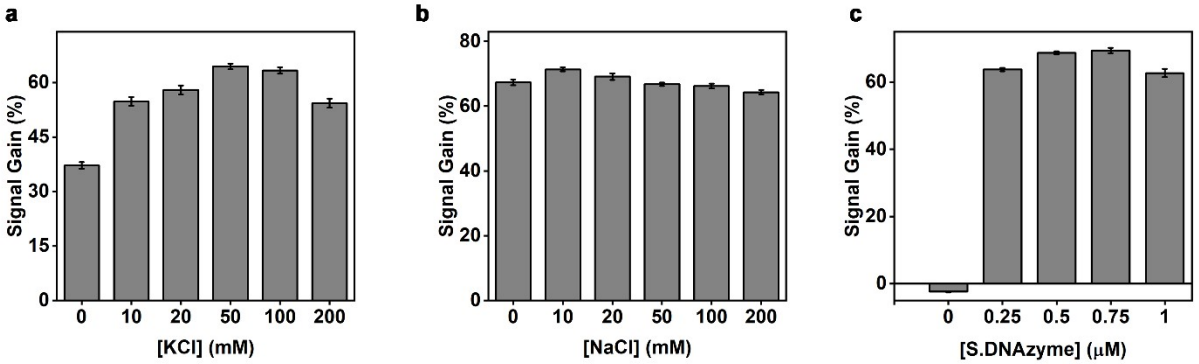

Fig. S3 Effects of (a) KCl, (b) NaCl and (c) S.DNAzyme concentration in reaction buffer on signal gain.

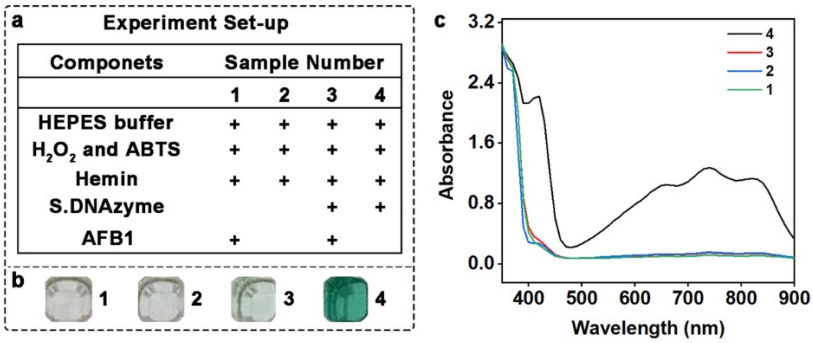

Fig. S4 Feasibility of S.DNAzyme for detection of AFB1. Experimental setup exhibits (a) sample contents, (b) photographs depict the color of the samples after 15 min of reaction and absorbance spectra of samples containing 1) reaction buffer with AFB1, 2) reaction buffer alone, 3) reaction buffer with S.DNAzyme and AFB1; 4) reaction buffer with S.DNAzyme (S.DNAzyme alone).

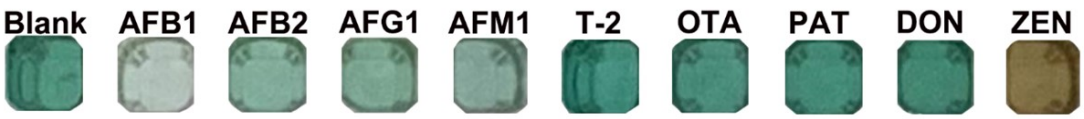

Fig. S5 Photographs of samples that contained various interferents (100 μM) after 15 minutes of reaction.

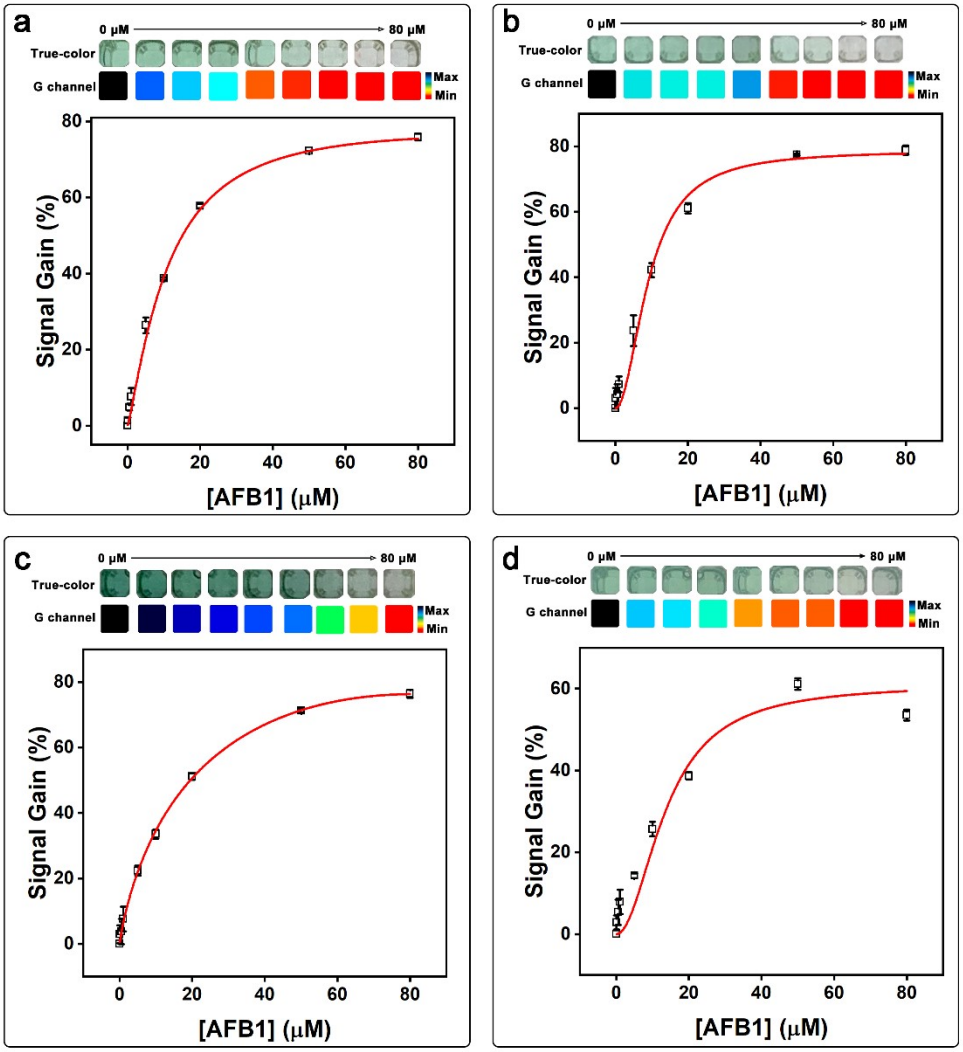

Fig. S6 Calibration curve produced using 0-80 μM AFB1 in traditional Chinese medicine (TCM) and food samples, including (a) adlay, (b) platyclade semen, (c) tangerine peel and (d) milk.

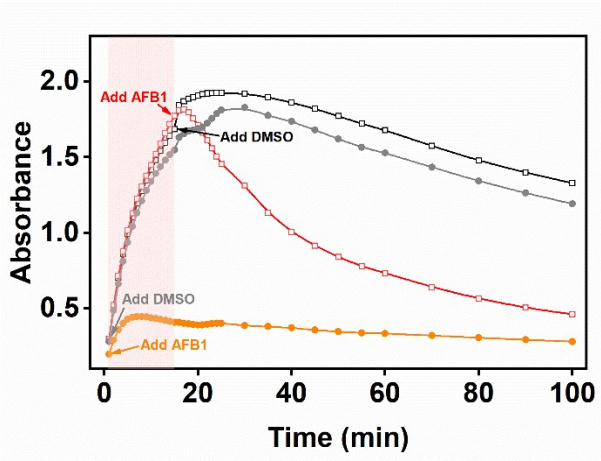

Fig. S7 Absorbance changes of samples at 418 nm during 0-100 min. AFB1 was added at 0 (yellow line) and 15 min (red line), respectively. DMSO was used as a control and was added into the solution at 0 (gray line) and 15 min (black line), respectively.

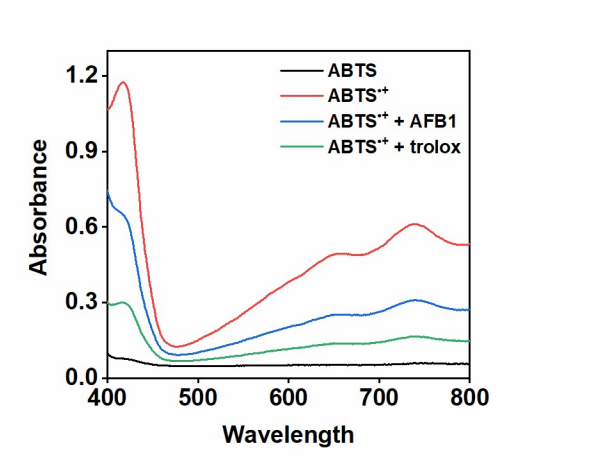

Fig. S8 UV-VIS absorption spectra of ABTS (black line), $ABTS^{\bullet+}$ (red line), $ABTS^{\bullet+}$ incubated with AFB1 ($ABTS^{\bullet+}$ + AFB1, blue line) and $ABTS^{\bullet+}$ incubated with trolox ($ABTS^{\bullet+}$ + trolox, green line).

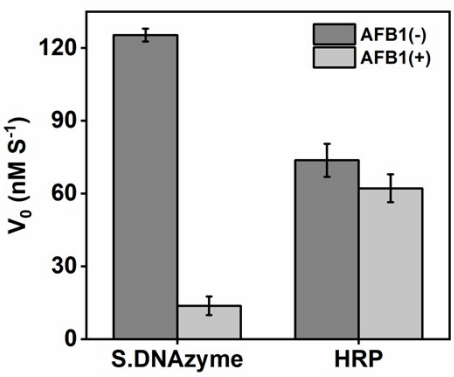

Fig. S9 Reaction rates of S.DNAzyme and HRP incubated without AFB1 (AFB1 (-)) or with AFB1 (AFB1 (+)) in terms of nanomolar $ABTS^{\bullet+}$ produced per second. The concentration of S.DNAzyme and HRP was 0.75 μM and 20 nM, respectively.

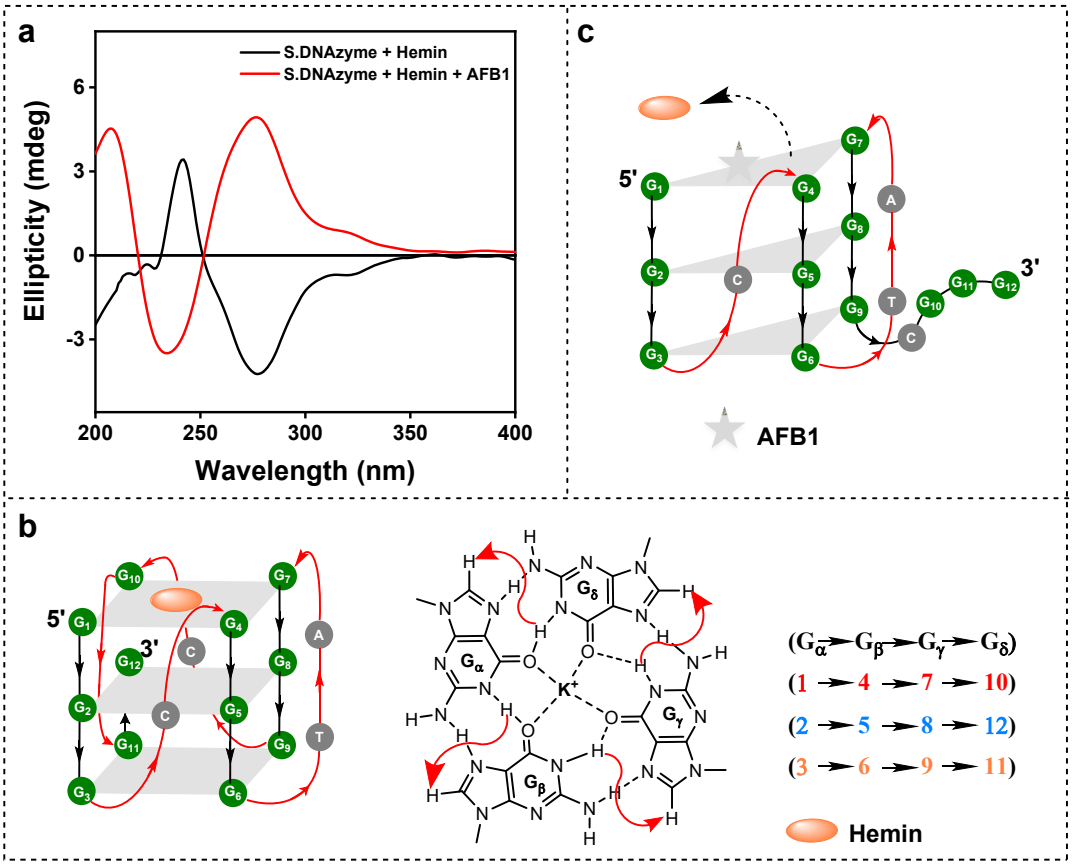

Fig. S10 (a) Circular dichroism spectra of S.DNAzyme with hemin (black) and S.DNAzyme with hemin with AFB1 (red). Schematics of the structure of S.DNAzyme with (b) hemin and (c) hemin and AFB1.

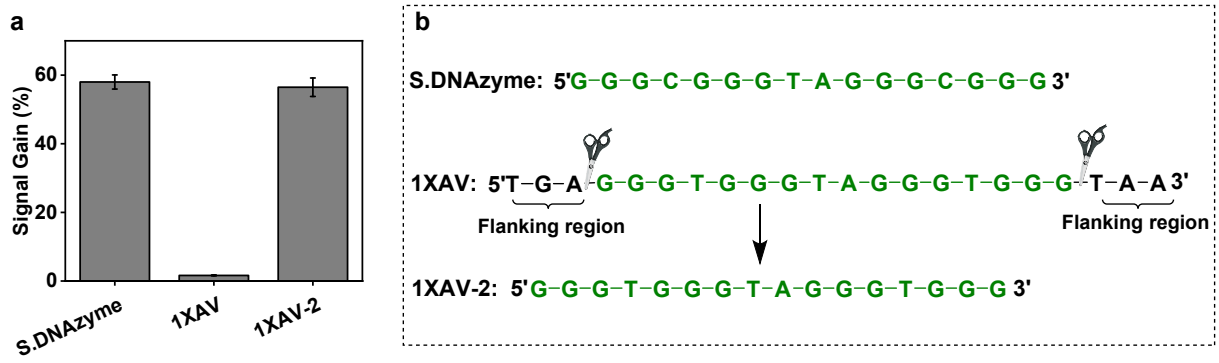

Fig. S11 (left) Signal gain and (right) sequences of AFB1 detection using S.DNAzyme, 1XAV and 1XAV-2.

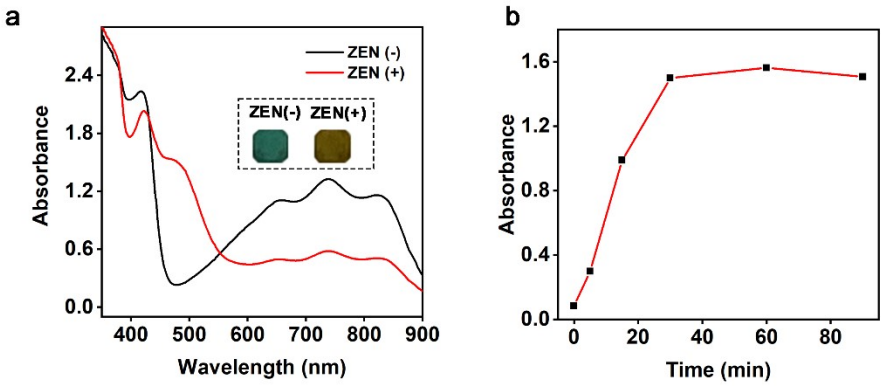

Fig. S12 (a) UV-VIS absorption spectra of the reaction solution without ZEN (ZEN (-)) and with ZEN (ZEN (+)), the illustration is the true color of solution after reaction for 15 minutes. (b) Absorbance changes at 472nm during 0 to 90 min.

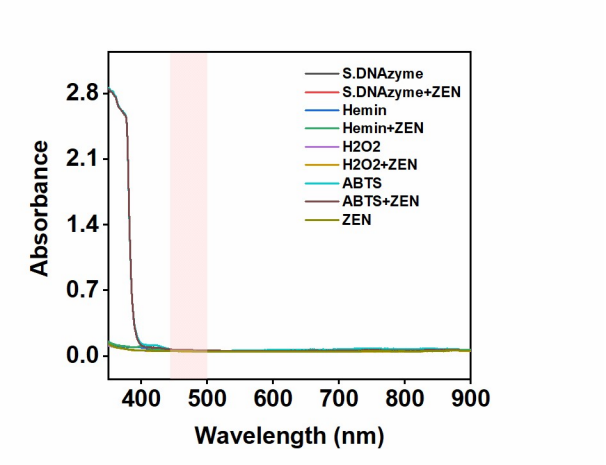

Fig. 13 The UV-VIS absorbance spectra of different solutions.

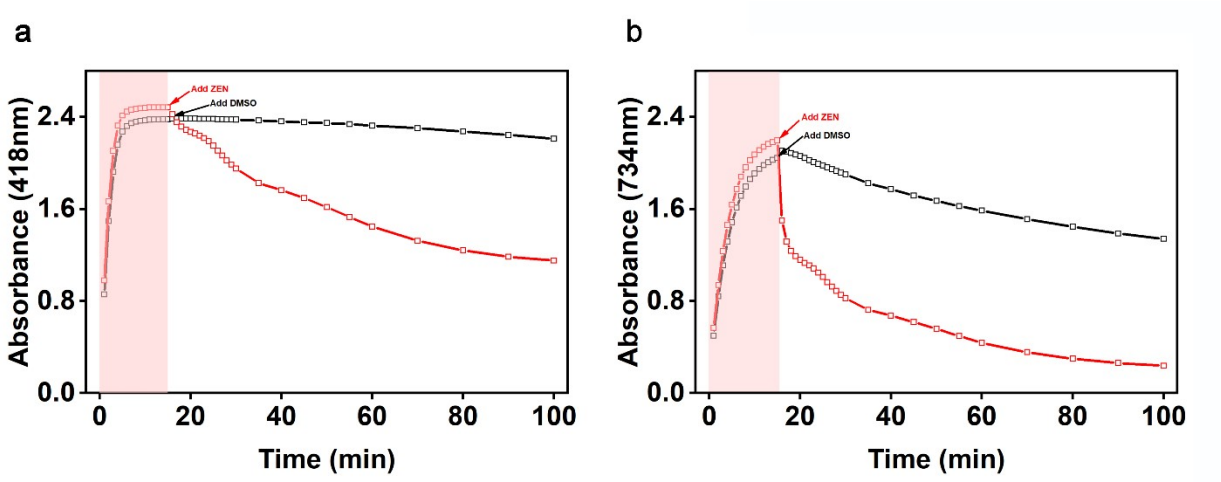

Fig. S14 Time-course absorbance of samples at (a) 418 nm and (b) 734 nm after adding ZEN or DMSO at 15 min. DMSO was used as control and was added to the blank solution.

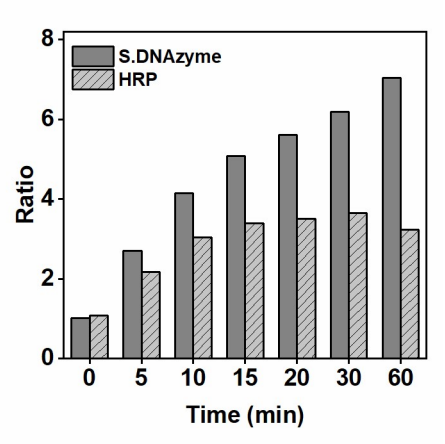

Fig. S15 The ratio change of ZEN detection at different time using S.DNAzyme and HRP. The ratio was calculated as follows: Ratio = absorbance value of experiment/ absorbance value of blank.

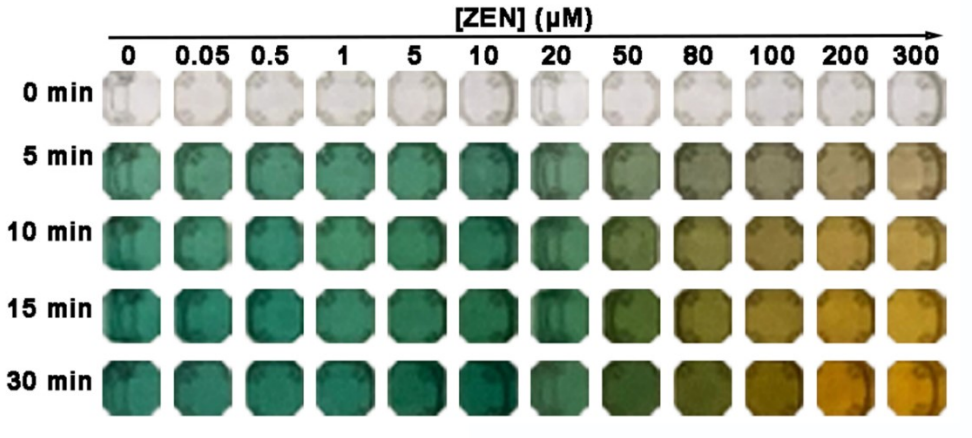

Fig. S16 The time course visual detection of ZEN using S.DNAzyme. Photographs of the samples containing different concentrations of ZEN (0-300 μM) at different time.

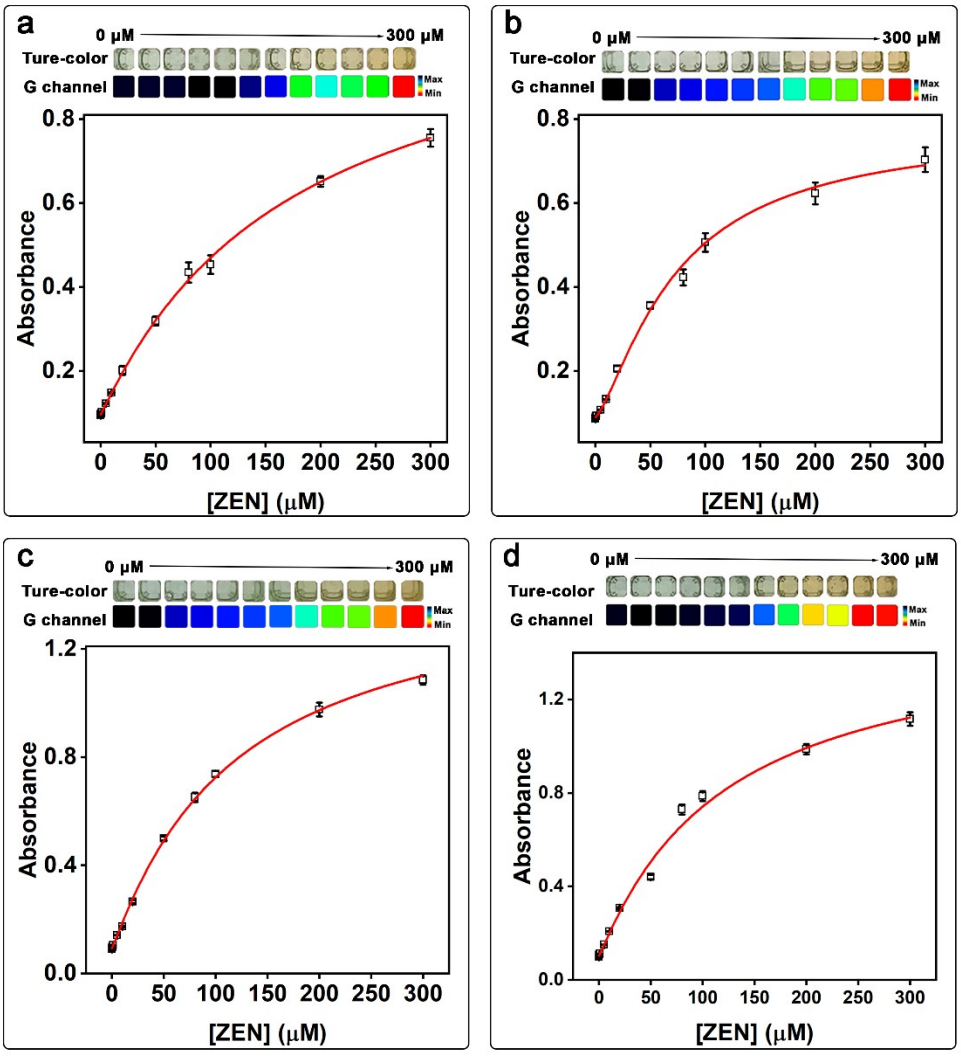

Fig. S17 Calibration curves of ZEN in real samples including (a) fritillariae cirrhosae bulbus, (b) angelica sinensis, (c) amygdalin and (d) rice.

References

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