Electronic Supplementary Material (ESI) for Nanoscale.
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Electronic Supplementary Material

Nanozyme-based cascade colorimetric aptasensor for amplified detection of ochratoxin A

Fengyu Tian [a,b,c], Jing Zhou [a,b,c], Bining Jiao [a,b,c] and Yue He [a,b,c*]

* To whom correspondence should be addressed. Dr. Yue He, E-mail: yuehe@cric.cn,

Tel: 86-23-68349603, Fax: 86-23-68349046

*Corresponding author:

[a] *Laboratory of Quality & Safety Risk Assessment for Citrus Products (Chongqing), Citrus Research Institute, Southwest University, Chongqing, 400712, P.R. China.*

[b] *National Citrus Engineering Research Center, Chongqing, P.R. China*

[c] *College of Food Science, Southwest University, Chongqing, 400712, P.R. China*

This file contains:

1. Supporting Discussion

2. Supporting Figures 1-8 with legends

1. Supporting Discussion

Optimization of assay conditions

The catalytic activity of nanozyme directly affects the sensitivity of this method. Thus, the MnO_2-catalyzed chromogenic reaction conditions including the concentrations of the MnO_2 nanosheets, pH of NaAc-HAc buffer, the concentrations of TMB, and the incubation time between MnO_2 nanosheets and TMB were optimized. The concentration of the MnO_2 nanosheets affected the signal of the aptasensor. As shown in Figure S4A, the absorbance intensity of MnO_2-TMB mixture increased rapidly with the increasing MnO_2 nanosheets concentration firstly and then decreased when the MnO_2 nanosheets concentration was higher than 13.44 μM. However, when the MnO_2 nanosheets concentration reached 13.44 μM, the absorbance intensity at 450 nm appeared obviously, which was 0.393 (Figure S4B) and the color of the solution became slightly blue-green (Figure S4C). This result demonstrates that an excess of MnO_2 nanosheets oxidized TMB to TMB^{2+}. As a result, 11.20 μM MnO_2 nanosheet was selected as the optimal concentration.

Then, the effect of the pH on the catalytic capacity of MnO_2 was studied. As can be seen from Figure S5, when pH 4.5 of NaAc-HAc buffer was used, the absorbance intensity of the MnO_2-TMB mixture reached the maximum value. Thus, pH 4.5 of NaAc-HAc buffer was used in the following experiments.

Since the concentration of TMB was an important parameter which might influence the absorbance intensity of MnO_2-TMB catalyst system. Thus, the effect of the concentration of TMB was investigated. As shown in Figure S6, it was observed that the absorbance intensity increased notably with the increasing concentration of TMB until 160 μM, where the absorbance plateaued. Therefore, 160 μM TMB was used.

The effect of incubation time between MnO_2 nanosheets and TMB on the absorbance intensity was studied. As shown in Figure S7, the absorbance intension increased progressively until 5 min and then reached a plateau. Hence, 5 min was chosen as the optimized incubation time.

Another important parameter, the concentration of AAP was optimized with a fixed concentration of OTA (12.5 μM). Figure S8 shows the effect of different AAP concentrations on the absorbance intensity. The absorbance intensity of MnO_2-TMB system decreased notably with the increasing concentration of AAP. 100 μM was chosen as the optimized condition.

2. Supporting Figures 1-8 with legends

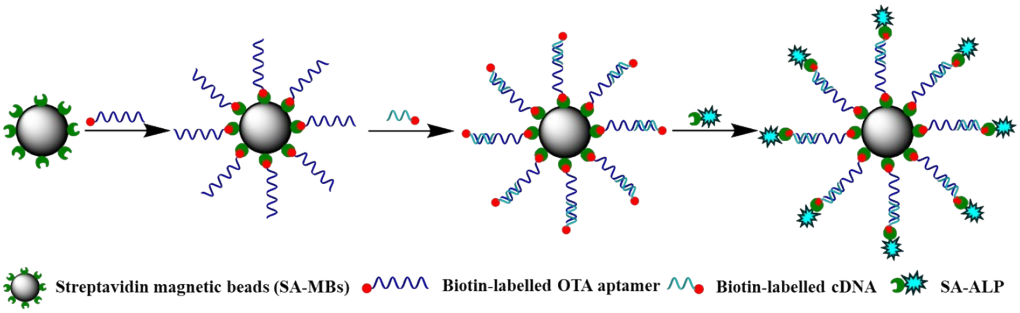

Figure S1 Preparation of DNA-ALP-immoblized MBs.

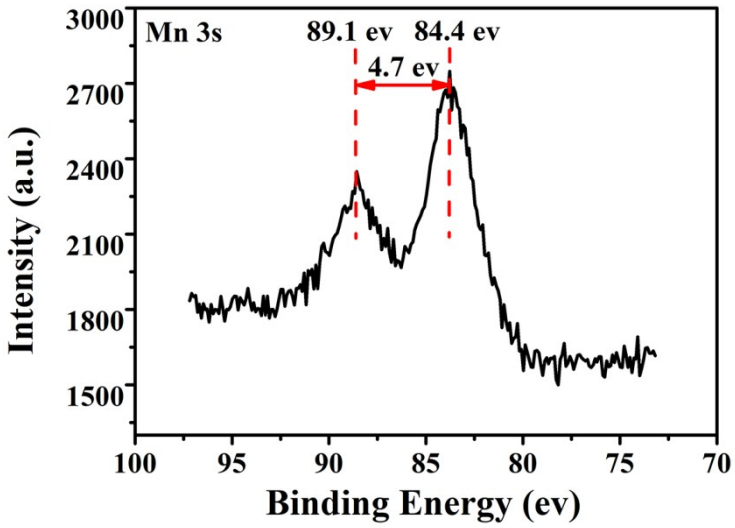

Figure S2 The high-resolution XPS pattern of Mn 3s.

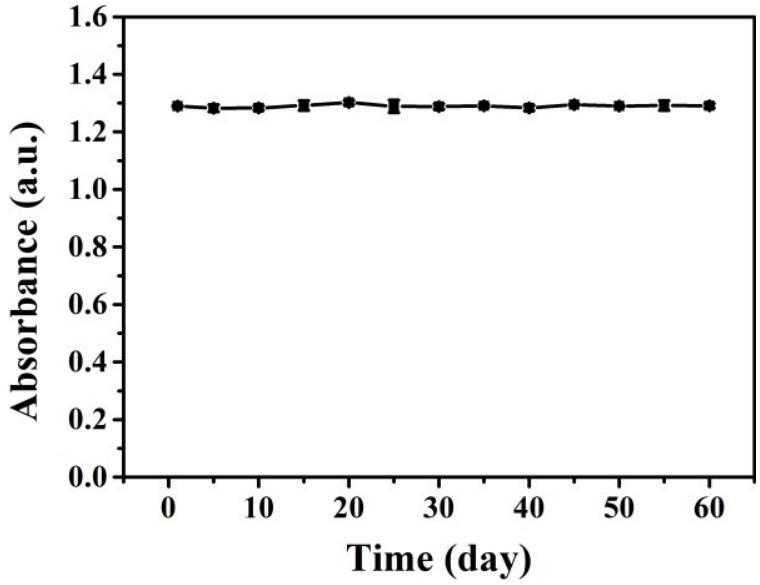

Figure S3 Stability of oxidase-like capacity of MnO_2 nanosheets.

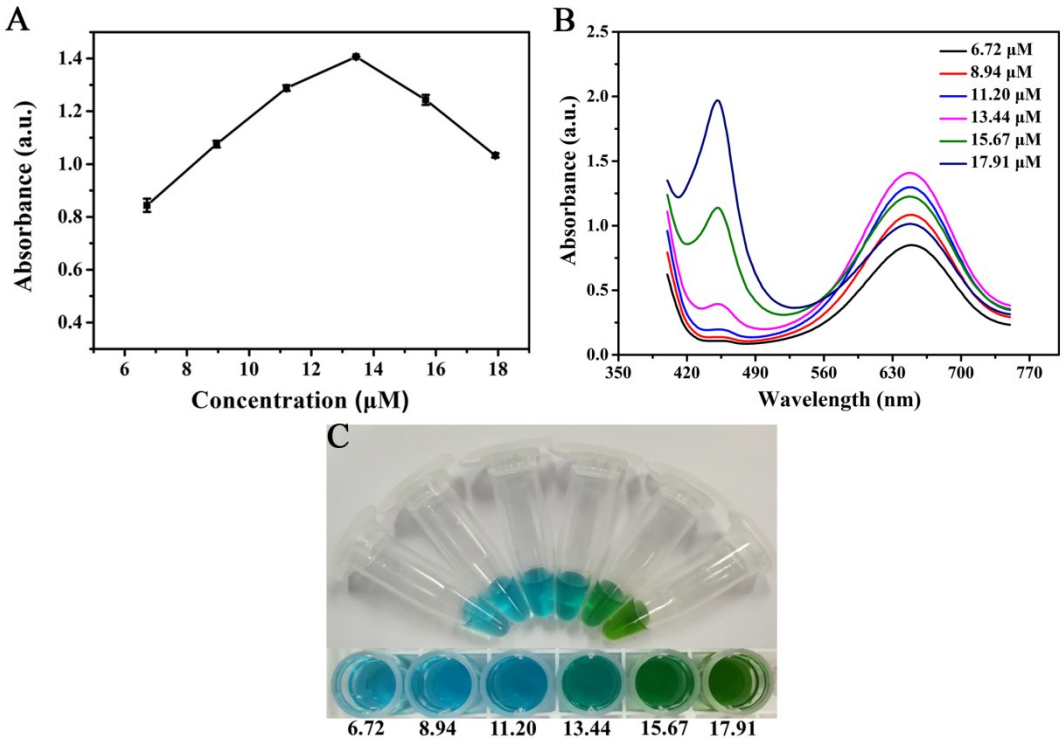

Figure S4 (A) The absorbance responses of the MnO_2-TMB system on addition of different concentrations of MnO_2 nanosheets. (B) UV-vis spectra of the MnO_2-TMB system following addition of various concentrations of MnO_2 nanosheets. (C) Corresponding photographs of the probe solution were taken under daylight. The error bars illustrate the standard deviations of three replicate measurements.

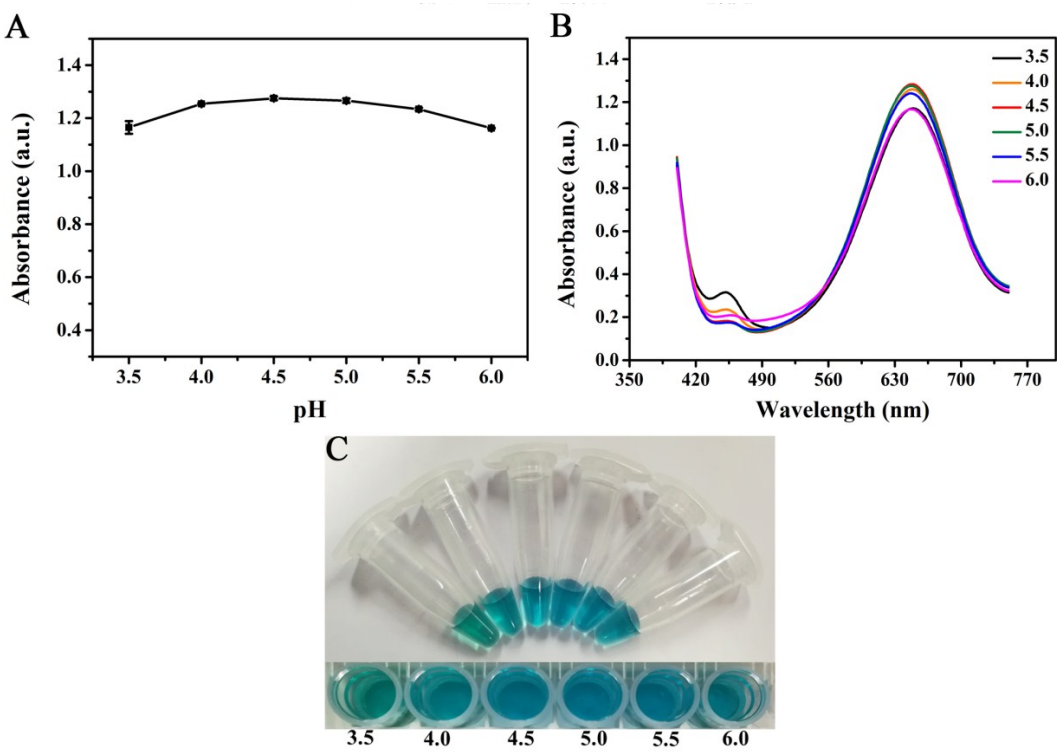

Figure S5 (A) The absorbance responses of the MnO_2-TMB system on various pH of NaAc-HAc buffer. (B) UV-vis spectra of the MnO_2-TMB system in various pH of NaAc-HAc buffer. (C) Corresponding photographs of the probe solution were taken under daylight. The error bars illustrate the standard deviations of three replicate measurements.

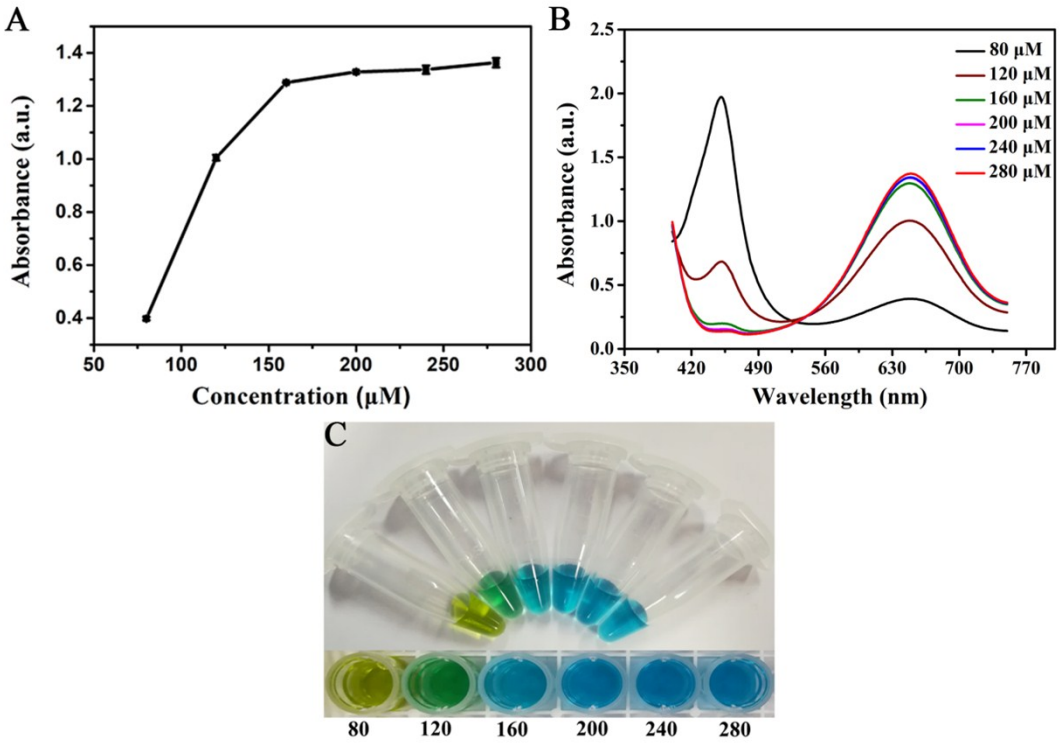

Figure S6 (A) The absorbance responses of the MnO_2-TMB system on addition of different concentrations of TMB. (B) UV-vis spectra of the MnO_2-TMB system following addition of various concentrations of TMB. (C) Corresponding photographs of the probe solution were taken under daylight. The error bars illustrates the standard deviations of three replicate measurements.

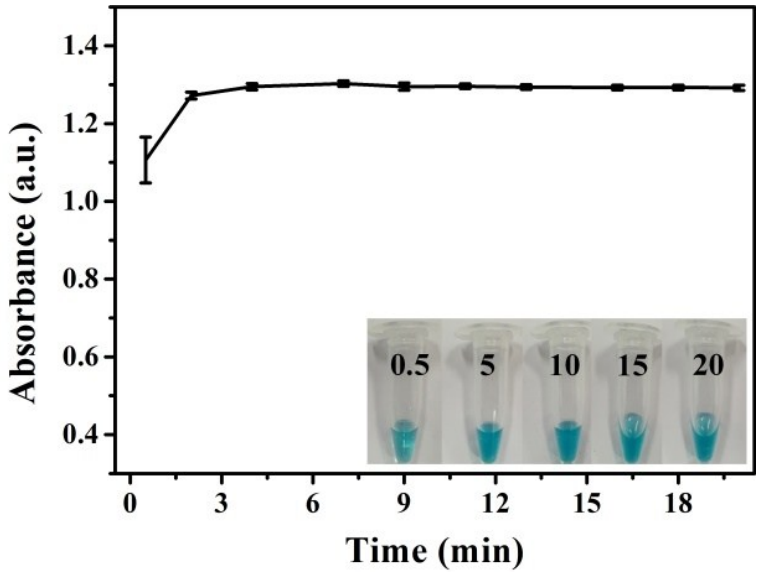

Figure S7 (A) The absorbance responses of MnO_2-TMB system under different incubation time. Insets are corresponding photographs of solution color. The error bars illustrate the standard deviations of three replicate measurements.

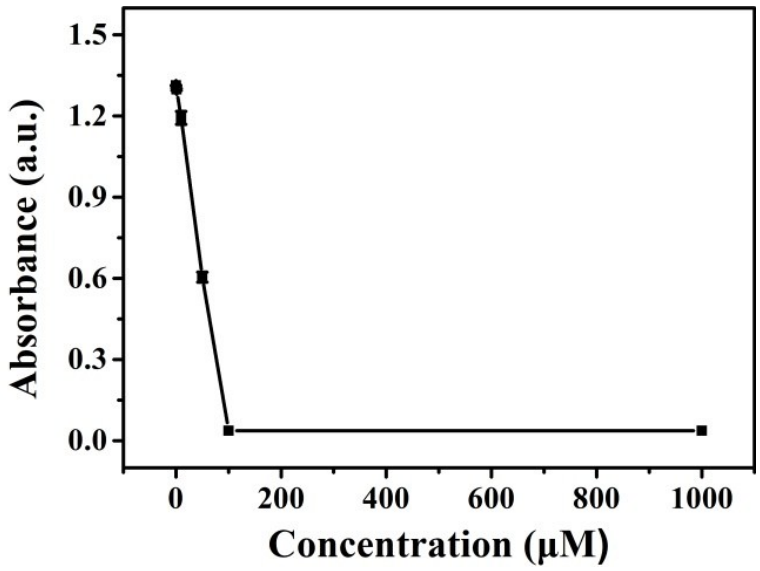

Figure S8 The absorbance responses of the ALP-MnO_2-TMB system on addition of different concentrations of AAP. The error bars illustrate the standard deviations of three replicate measurements.