

## Electronic Supplementary Material

*for*

### **Smartphone-based colorimetric assay of antioxidants in red wine using oxidase-mimic MnO<sub>2</sub> nanosheets**

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## Experimental

### Chemicals and Materials

Manganese chloride tetrahydrate ( $\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$ ), tetramethylammonium hydroxide ( $\text{TMA} \cdot \text{OH}$ ), 3,3',5,5' - tetramethylbenzidine dihydrochloride (TMB), gallic acid (GA), L-ascorbic acid (AA), tannic acid (TA), and catechol were all obtained from Aladdin Industrial Corporation (Shanghai, China). Hydrogen peroxide ( $\text{H}_2\text{O}_2$ , 30 wt%) was purchased from Chuandong Chemical Co., Ltd (Chongqing, China). All the chemicals were used as received. NaAc-HAc buffer was used to control the acidity of the solution in detection. Milli-Q purified water ( $18.2 \text{ M}\Omega \cdot \text{cm}$ ) was used throughout the experiments.

### Instrumentations

The absorption spectra of  $\text{MnO}_2$  nanosheets and oxTMB were measured using a UV-2550 UV-vis spectrophotometer (Shimadzu, Japan). Transmission electron microscopy (TEM) images of the nanosheets were taken using a Tecnni G2 F20 transmission electron microscopy (USA). Dynamic light scattering (DLS) data of  $\text{MnO}_2$  nanosheets were taken using a Brookhaven Nano Brook omni (USA). A high-speed TGL-16 M centrifuge (Hunan, China) was used in the purification of samples. A numerical controlled KH-2200DE ultrasonic cleaner (Shandong, China) was used to disperse the  $\text{MnO}_2$  nanosheets in solution.

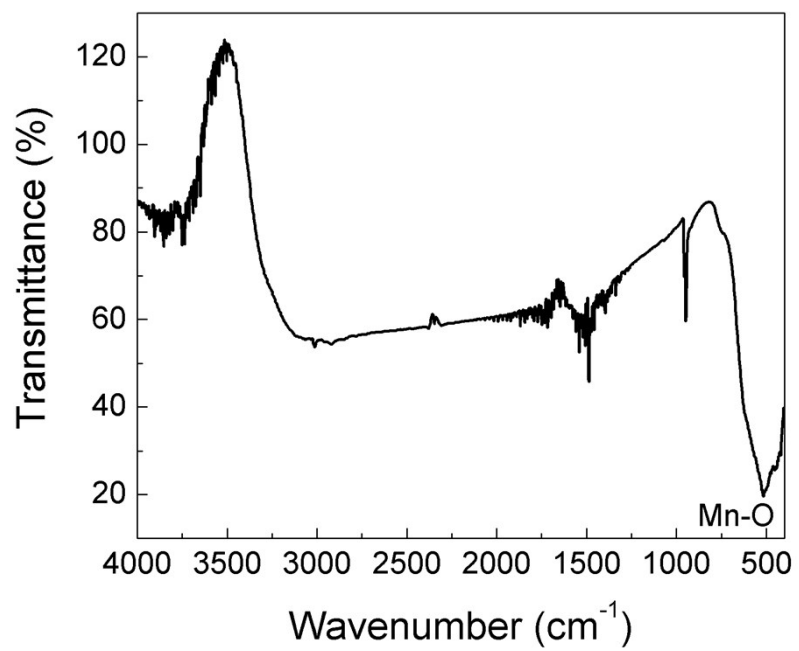
### Synthesis of $\text{MnO}_2$ nanosheets

$\text{MnO}_2$  nanosheets were prepared according to previously reported protocol with slight modification.<sup>1</sup> Briefly, an aqueous solution (20 mL) containing 0.6 M TMA·OH and 3 wt%  $\text{H}_2\text{O}_2$  was prepared firstly. Then, this solution was rapidly mixed with 10 mL  $\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$  (0.3 M). The mixture became dark brown immediately, confirming that  $\text{Mn}^{2+}$  was oxidized to  $\text{Mn}^{4+}$ . The obtained dark brown solution was stirred vigorously overnight at room temperature. The precipitate was obtained by centrifugation at 10000 rpm for 10 min and washed three times with water and methanol. Finally, bulk  $\text{MnO}_2$  was obtained by drying the precipitates in an oven at  $30^\circ\text{C}$ .

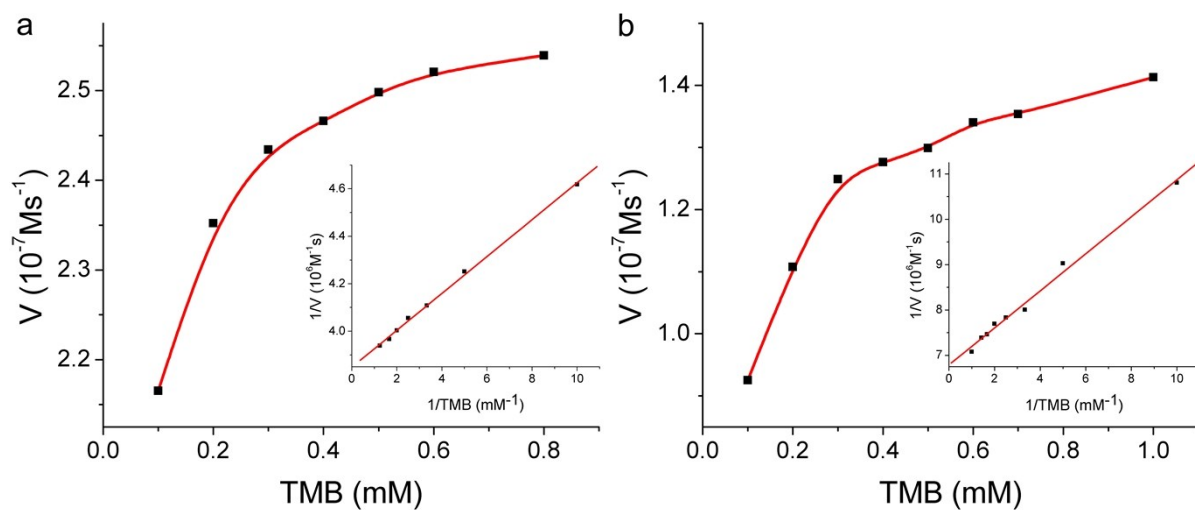
To obtain  $\text{MnO}_2$  nanosheets in solution, 10 mg bulk  $\text{MnO}_2$  was added to 100 mL water and ultrasonicated for 3h. After this process, the suspension was centrifuged at 2000 rpm for 10 min to remove the unexfoliated  $\text{MnO}_2$ . The supernatant was collected and stored in refrigerator at  $4^\circ\text{C}$  for further use.

### **Procedure for the detection of antioxidants**

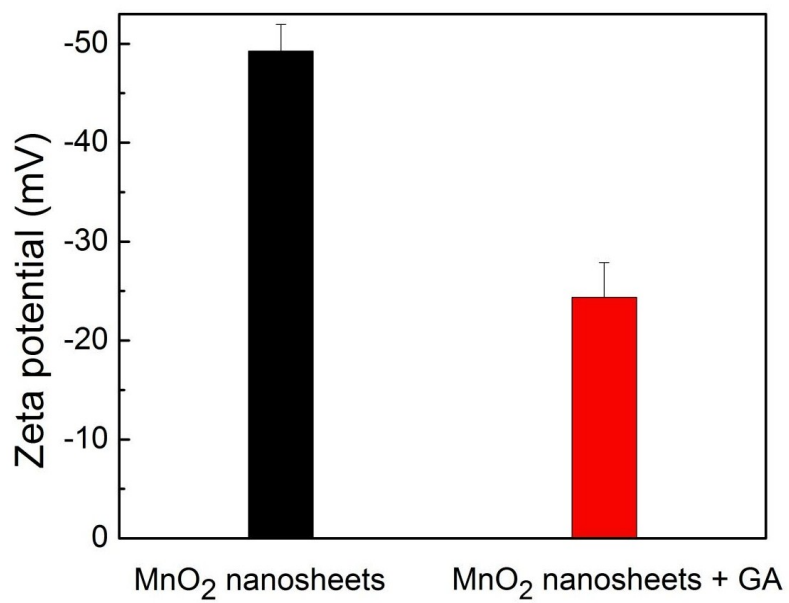
In a standard procedure, 800  $\mu\text{L}$  of standard solution of antioxidant (e.g., GA) with an appropriate concentration was mixed with 800  $\mu\text{L}$  of  $\text{MnO}_2$  nanosheets (0.088 mM). After reaction for 5 min at room temperature, 800  $\mu\text{L}$   $\text{H}_2\text{O}$ , 800  $\mu\text{L}$  NaAc-HAc buffer (pH 4.8) and 800  $\mu\text{L}$  TMB (0.4 mM) were added to this solution sequentially. UV-vis spectra measurement of the resulting solution was carried out after 10 min. For the assay using a smartphone, the photograph was first taken by the phone (iPhone 7 with an iOS 10 operating system) after reaction for 10 min. Then, the samples in the photograph were analyzed by an open-source app called *Color Pickers*. The RGB parameters of each sample were obtained as signals for assay. In order to evaluate the practicability of this method, the antioxidants in red wine were also determined by the same protocol as that of the standard, where the red wine was only processed by dilution with water.



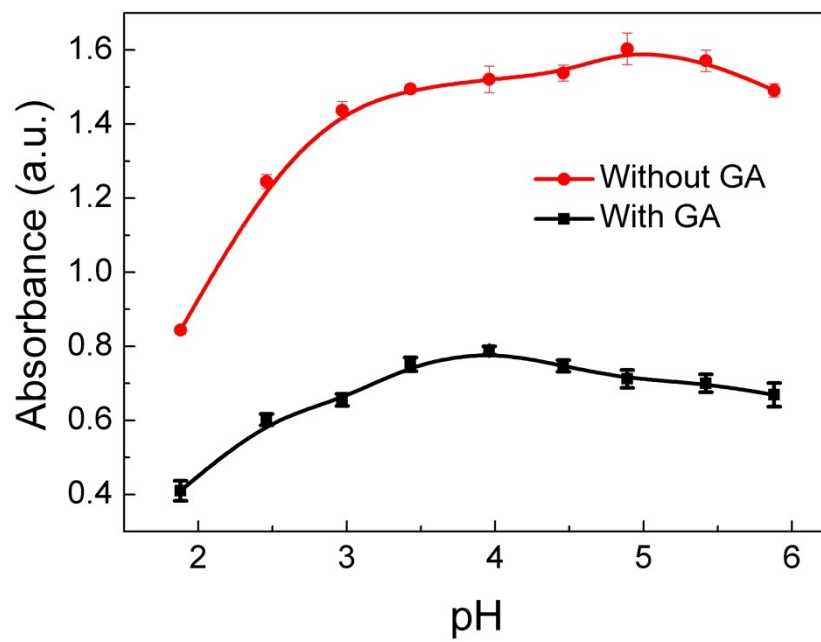
**Fig. S1** The Fourier transform infrared (FT-IR) spectrum of MnO<sub>2</sub> nanosheets.



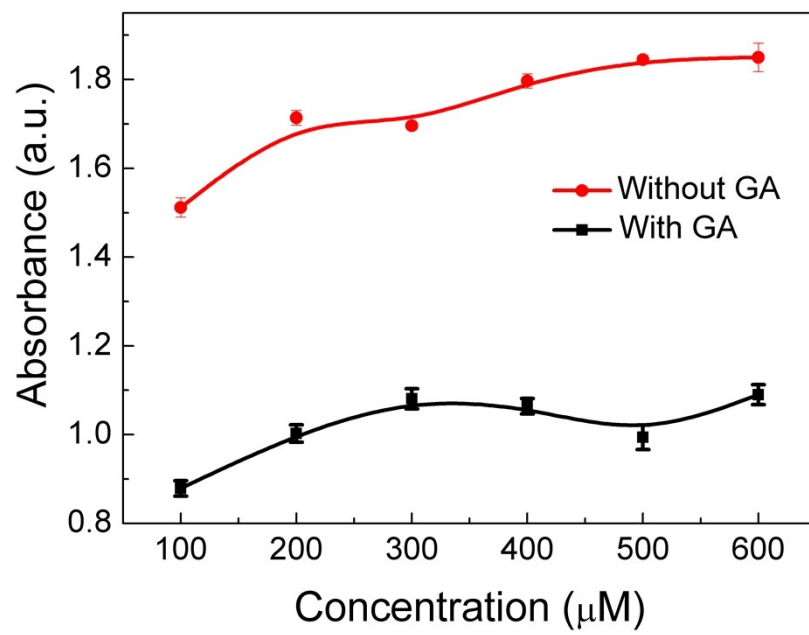
**Fig. S2** The Steady-state kinetic assays of the  $\text{MnO}_2$  nanosheets in the absence (a) and presence (b) of GA. Insets are the Lineweaver-Burk plots of the Michaelis-Menten equations.



**Fig. S3** The apparent zeta potential values of MnO<sub>2</sub> nanosheets before and after the addition of GA.

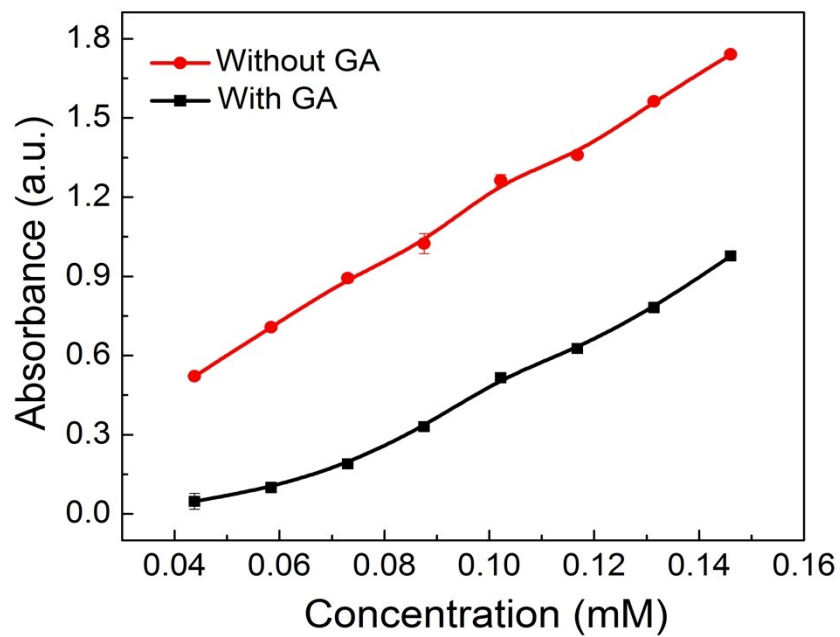


**Fig. S4** The effect of solution pH on the detection of antioxidants.



**Fig. S5** The effect of TMB concentration on the detection of antioxidants.

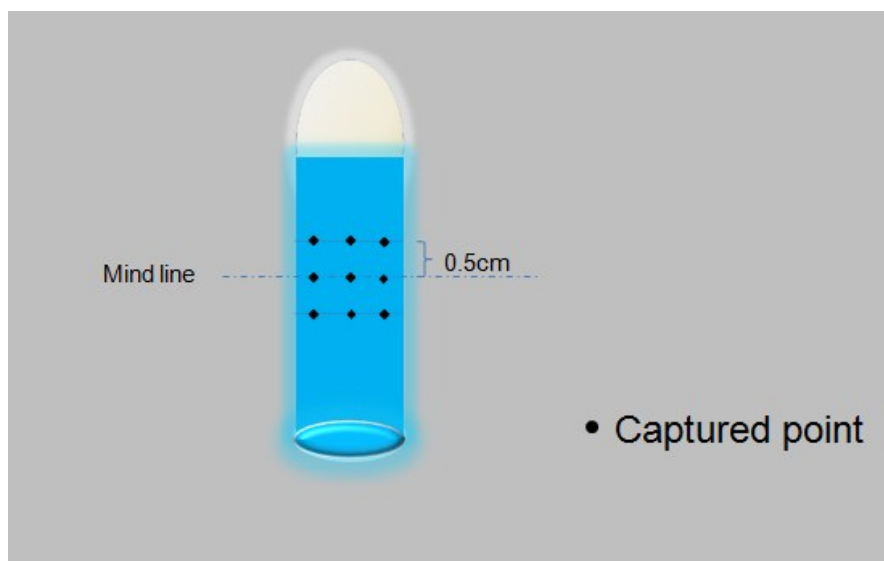




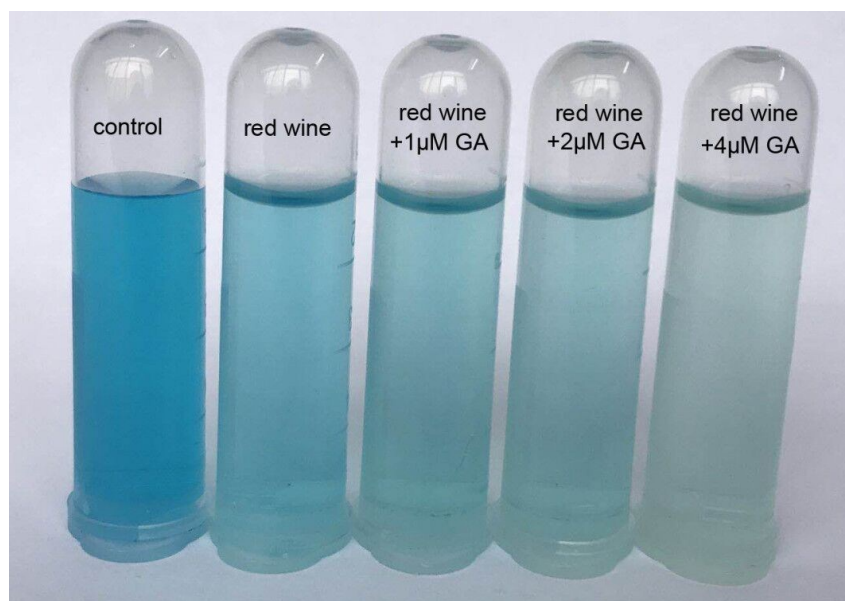
**Fig. S6** The effect of MnO<sub>2</sub> nanosheets concentration on the detection of antioxidants.



**Fig. S7** Screen captures during the antioxidant detection by a smartphone: (a) iPhone menu loaded app, (b) main menu of the *Color Pickers* app, (c) typical sample analysis process, and (d) final results in RGB mode obtained after image processing.



**Fig. S8** Schematic illustration showing the typical sample sites of captured points by *Color Pickers*.



**Fig. S9** Photograph of the sample solutions in standard addition experiments (from left to light: control, red wine, red wine with 1  $\mu\text{M}$  GA , red wine with 2  $\mu\text{M}$  GA, and red wine with 4  $\mu\text{M}$  GA).

**Table S1** Comparison of the kinetic parameters between horse radish peroxidase (HRP) and MnO<sub>2</sub> nanosheets.  $K_m$  is the Michaelis-Menten constant and  $V_{max}$  is the maximal reaction rate.

Catalyst	Substrate	$K_m$ / mM	$V_{max}$ / M s <sup>-1</sup>	Reference
HRP	TMB	0.434	$1.00 \times 10^{-7}$	2
MnO <sub>2</sub> nanosheets	TMB	0.0114	$2.60 \times 10^{-7}$	this work
MnO <sub>2</sub> nanosheets + GA	TMB	0.106	$1.47 \times 10^{-7}$	this work

**Table S2** The linear equation, correlation coefficient ( $r$ ), and detectable range of the RGB methods for antioxidant detection (in term of GA equivalents).

	R method	G method	B method
equation	$I_R = 10.60c_{GA} + 3.672$	$I_G = 4.807c_{GA} + 116.1$	$I_B = 3.177c_{GA} + 149.2$
$r$	0.9847	0.9931	0.9917
liner range	3~15 $\mu$ M	3~15 $\mu$ M	3~15 $\mu$ M

**References:**

1. K. Kai, Y. Yoshida, H. Kageyama, G. Saito, T. Ishigak, Y. Furukawa, J. Kawamata, *J. Am. Chem. Soc.*, 2008, **130**, 15938–15943.
2. J. Wei, X. Chen, S. Shi, S. Mo, N. Zheng, *Nanoscale*, 2015, **7**, 19018–19026.