Electronic Supplementary Material

A CRISPR/Cas12a-based Fluorescent Method for the Amplified Detection of Total Antioxidant Capacity

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1. Supporting figures 1-6 with legends

2. Supporting tables 1-3

1. Supporting figures 1-5 with legends

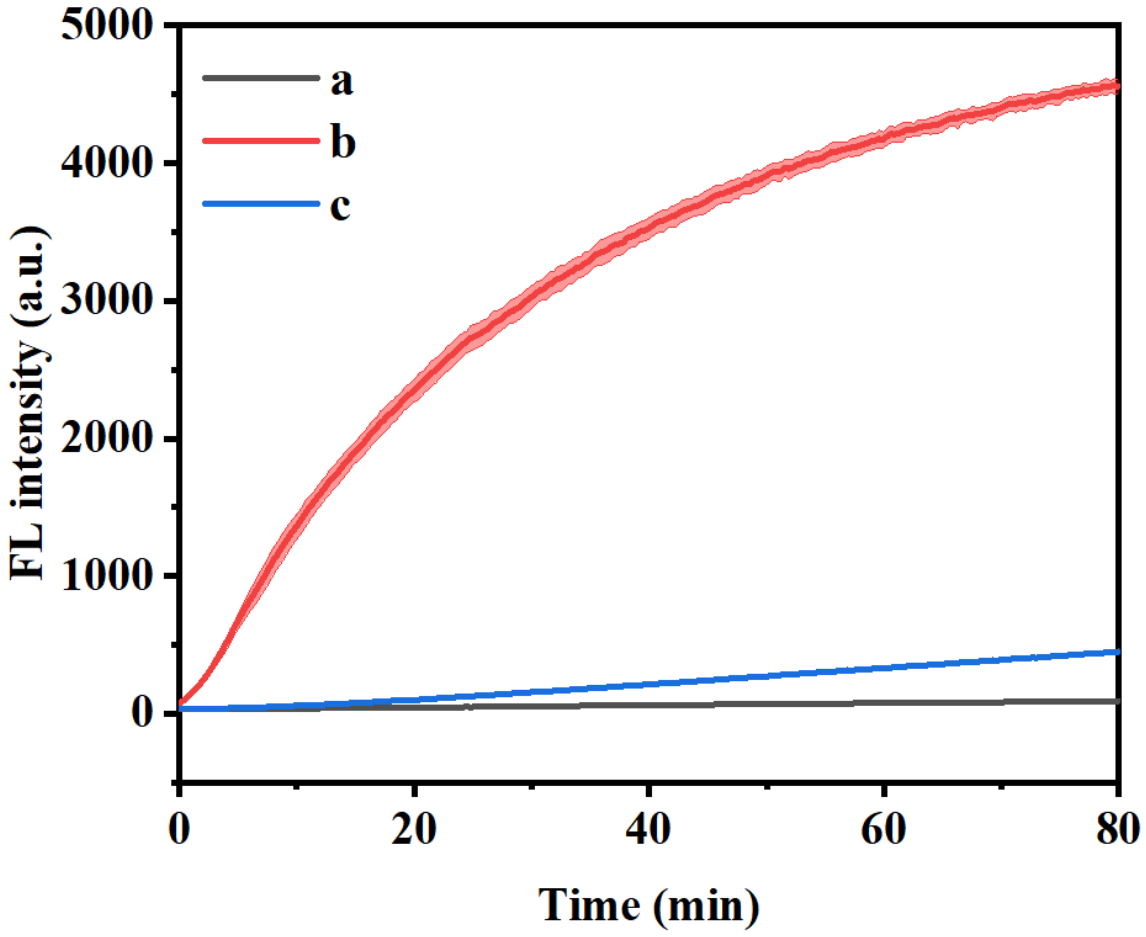

Fig. S1 Time-dependent *trans*-cleavage signals of Cas12a under different treatments: (a) CRISPR/Cas12a system; (b) CRISPR/Cas12a system + Mn^{2+}; (c) CRISPR/Cas12a system + (Mn^{2+} + EDTA). The CRISPR/Cas12a system contain Cas12a-crRNA complex (25 nM), substrate ssDNA (20 nM), and ssDNA-FQ (500 nM). Mn^{2+}, 30 μM; EDTA, 30 μM

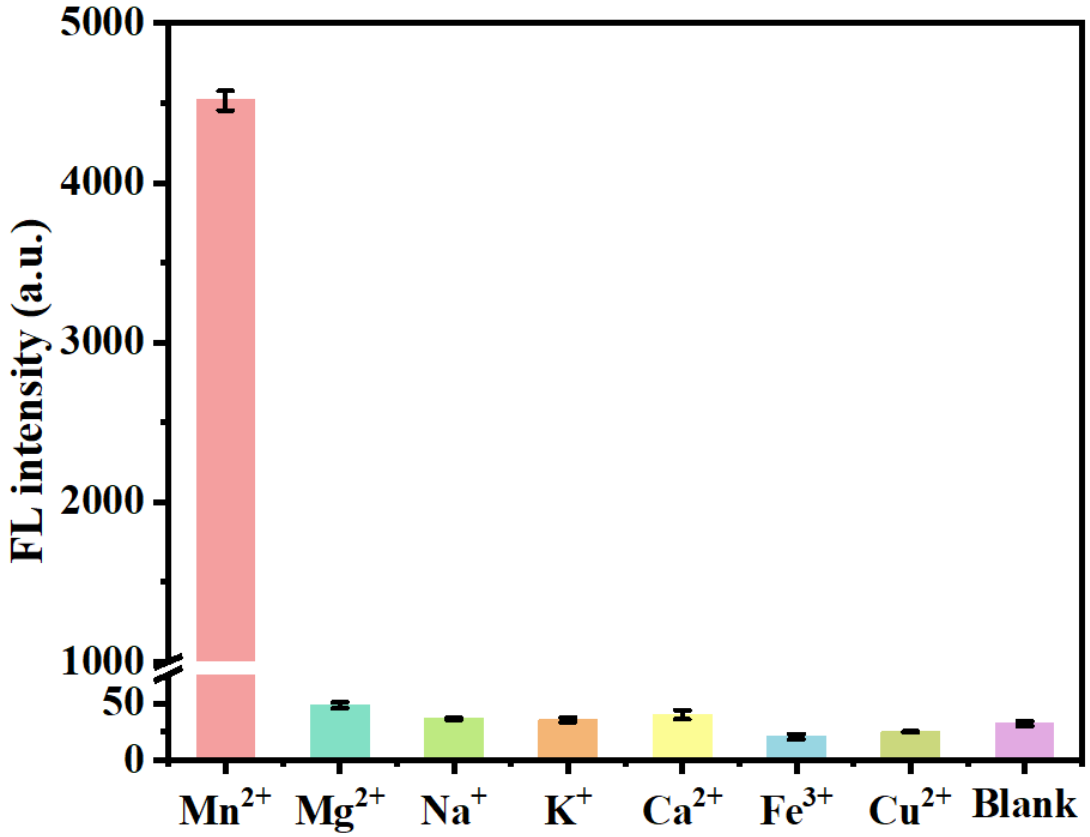

Fig. S2 Assess the effect of metal ions on the *trans*-cleavage activity of Cas12a. The concentrations of Mn^{2+} and Mg^{2+} were 30 μM. And the concentrations of other metal ions were 60 μM. Cas12a-crRNA complex, 25 nM; ssDNA-FQ, 500 nM; substrate ssDNA, 20 nM. The error bars were calculated using the standard deviation (SD) of three repeated experiments

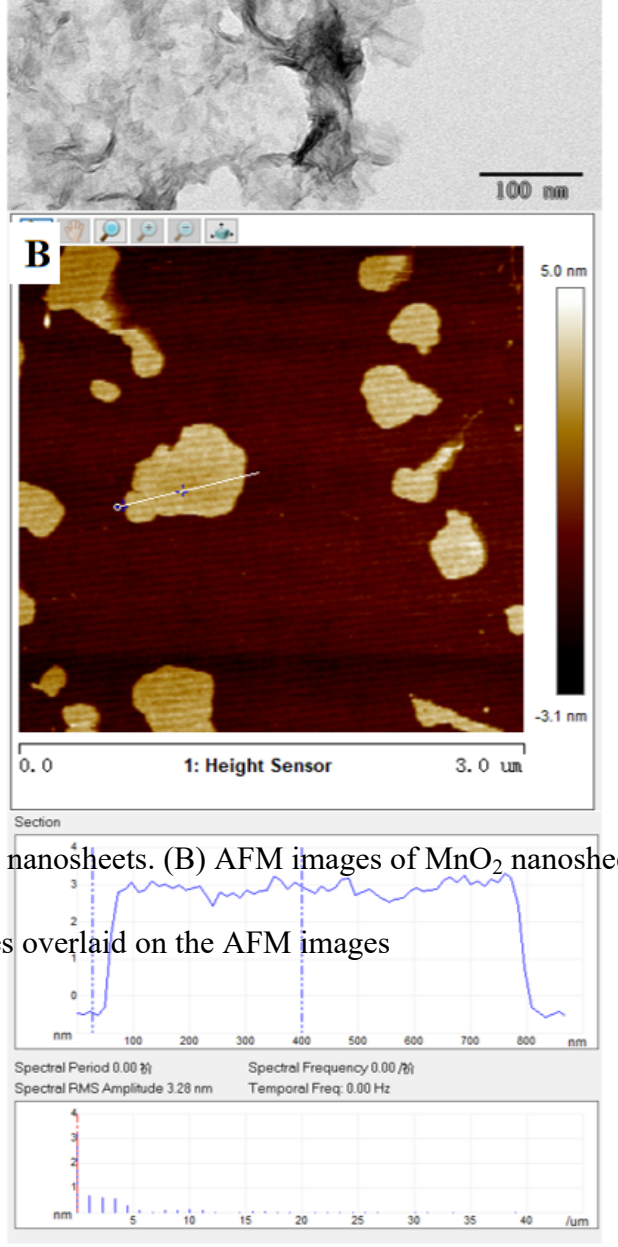

Fig. S3 (A) TEM images of MnO_2 nanosheets. (B) AFM images of MnO_2 nanosheets and height profiles along the white lines overlaid on the AFM images

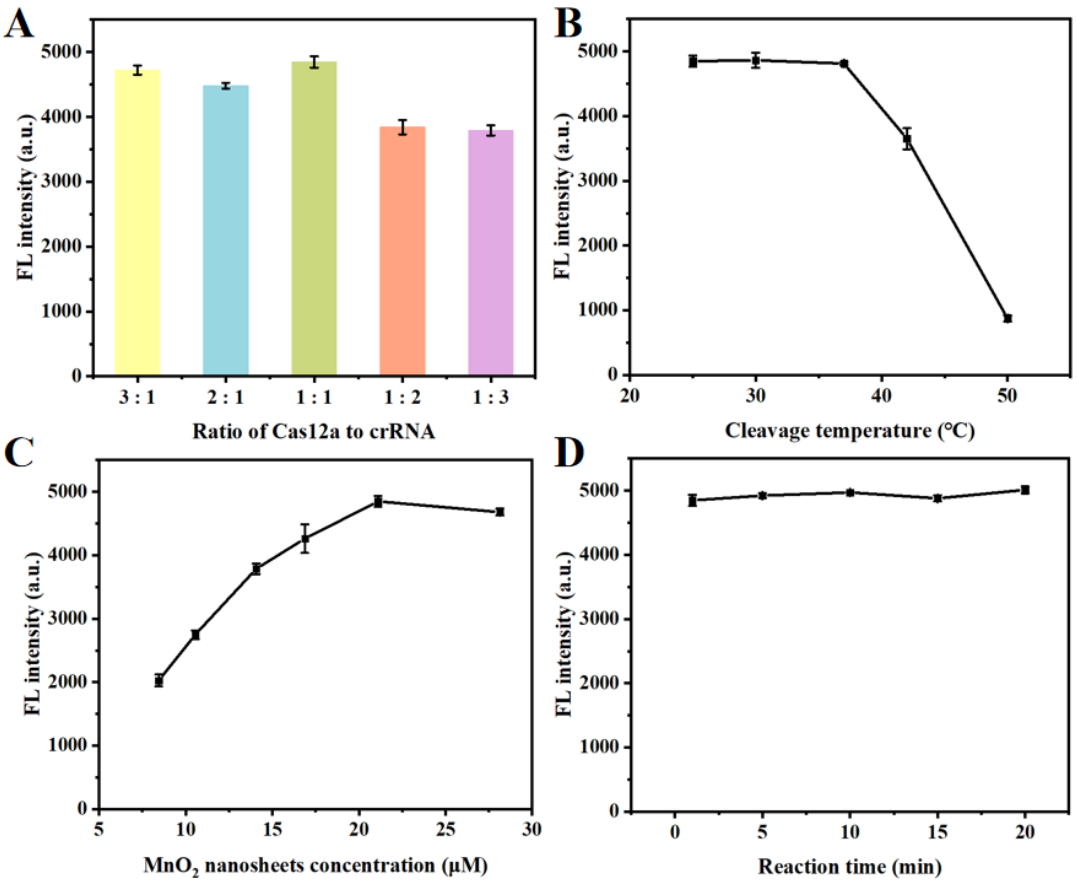

Fig. S4 Conditions optimization of the proposed FL method. (A) The molar ratio of Cas12a to crRNA. (B) The cleavage temperatures for Cas12a. (C) The concentration of MnO_2 nanosheets. (D) The reaction time between AA and MnO_2 nanosheets. Cas12a-crRNA complex, 25 nM; ssDNA-FQ, 500 nM; substrate ssDNA, 20 nM; AA, 25 μM; MnO_2 nanosheets, 21.1 μM. The error bars were calculated using the SD of three repeated experiments

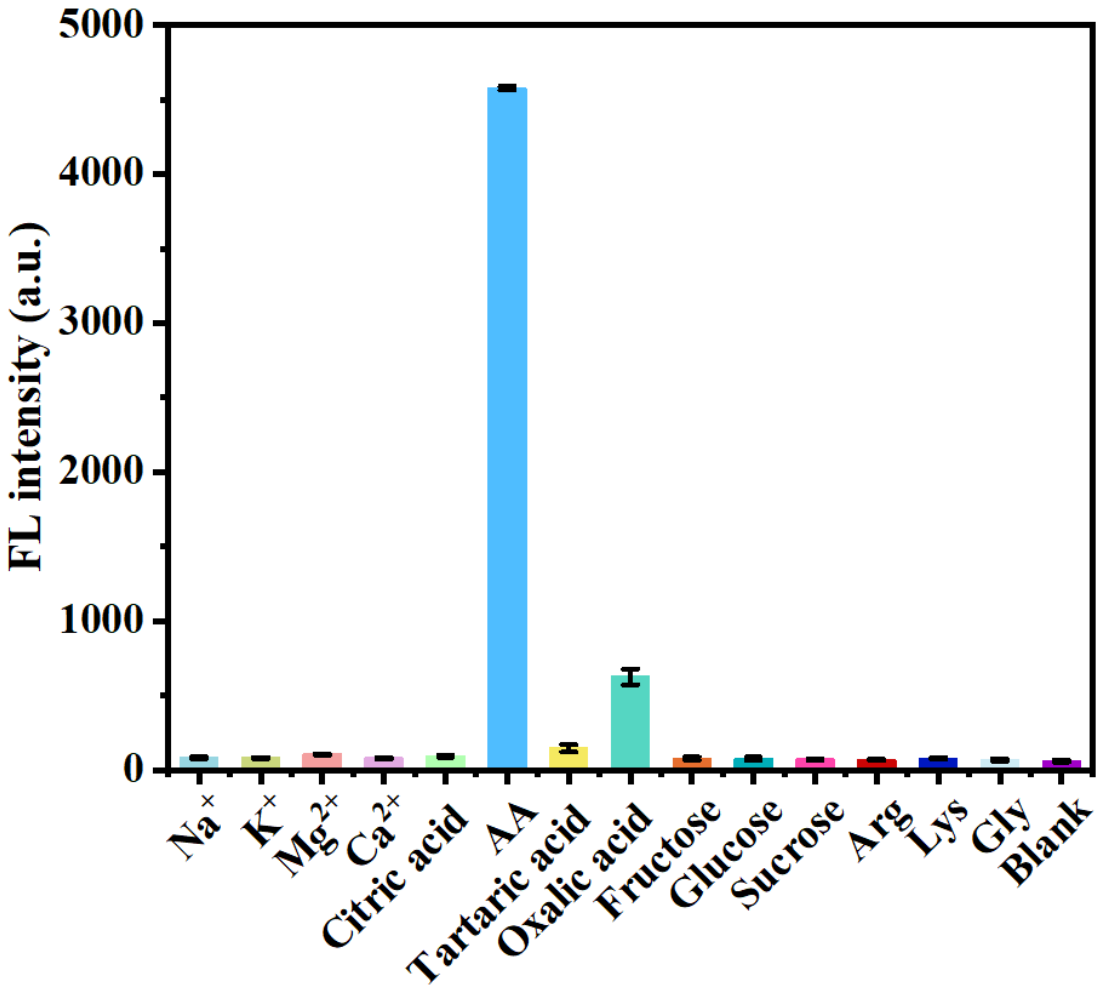

Fig. S5 The FL response intensities of the proposed FL method to Na^+, K^+, Mg^{2+}, Ca^{2+}, citric acid, tartaric acid, oxalic acid, fructose, glucose, sucrose, Arg, Lys, Gly and AA at a concentration of 20 $mg \cdot L^{-1}$. The error bars were calculated using the SD of three repeated experiments

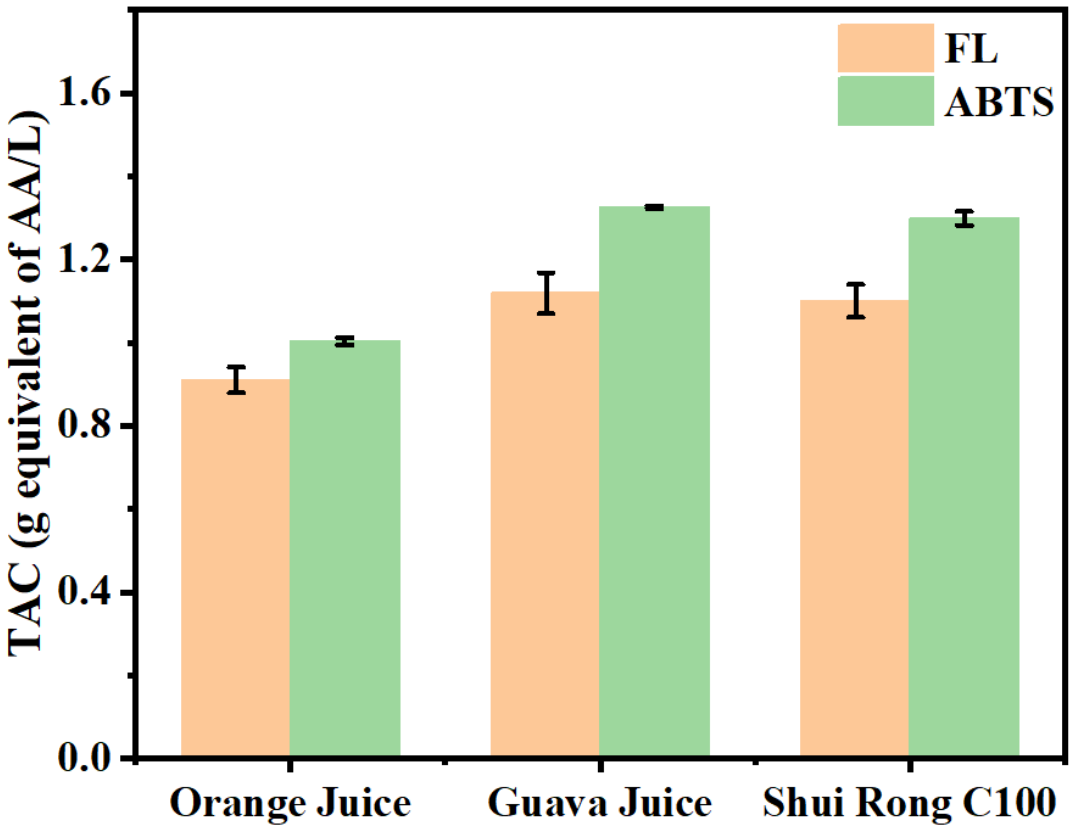

Fig. S6 Determined TAC values in the three beverage samples and comparison with standard ABTS assay

2. Supporting tables 1-3

Table S1 Oligonucleotide sequences used in this work

Oligonucleotide	Sequence（5′→3′）
crRNA	UAAUUUCUACUAAGUGUAGAUUCCUAGUGGUGGGCGAACCC
substrate ssDNA	GGGTTCGCCCACCACTAGGA
ssDNA-FQ	FAM-TTATT-BHQ1

Table S2 The FL intensity of three repeated experiments and the corresponding relative standard deviation (RSD) values of the sensing solution for different concentrations of AA

Concentration ($mg \cdot L^{-1}$)	**Data**			**Average value**	**RSD/%**
20	4595	4466	4646	4569.00	2.03
15	4358	4181	4501	4346.67	3.69
12.5	4146	4156	4051	4117.67	1.41
10	3149	3206	3123	3159.33	1.34
7.5	2416	2382	2579	2459.00	4.28
5	1365	1107	1120	1197.33	12.14
2.5	375.2	483.5	398.5	419.07	13.60
1	175.4	157.4	136.8	156.53	12.34
0.5	72.97	97.02	78.41	82.80	15.23
0.1	54.17	63.25	54.58	57.33	8.94
0	52.02	60.54	58.57	57.04	7.82
Average value					7.53

Table S3 Comparison the analytical performance of different fluorescent methods for AA detection

Materials	Linear Range (μM)	Detection Limit (μM)	Reference
BSA-AuNCs	3-50	0.4	[1]
CrO_4^{2-}@Cd-MOFs	46.3-591	41.28	[2]
GQDs–hypochlorite	8-60	1.4	[3]
RhB@MOF nanocomposite	1-25	0.31	[4]
CuInZnS QDs@FeOOH	5–60	1.5	[5]
DBHM + Cu^{2+} sensor	0-500	2.37	[6]
Cu NPs/N–$Ti_3C_2T_x$	5-150	0.437	[7]
NiNCs/Fe^{3+} composites	20–200	7.45	[8]
CA-CDs	5-100	0.15	[9]
Fe_3O_4@SiO_2@UiO-PBA	5–60	2.5	[10]
CRISPR/Cas12a-MnO_2	2.84-70.97	0.23	This work

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