Supplementary Information

Guanosine-rich Aptamers@Cu_2O Nanoparticles: Enhanced Peroxidase Activity and Specific Recognition Capability at Neutral pH

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Experimental procedure

1.1 Materials

Copper (II) chloride dihydrate and ascorbic acid were obtained from Aladdin. Sodium hydroxide and hydrogen peroxide were purchased from Shanghai Chemical Reagent Co., Ltd. The 3,3',5,5'-tetramethylbenzidine (TMB), 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS), thrombin, bovine serum albumin, human IgG and lysozyme were bought from Sigma-Aldrich. All ssDNA sequences were synthesized and purified by Sangon Biotech Co., Ltd.

1.2 Preparation of hollow Cu_2O nanocubes

Firstly, about 120 μL of 0.1 M $CuCl_2$ aqueous solution and 38 μL of 0.6 M NaOH aqueous solution were sequentially added into 2.0 mL water. Then 90 μL of 0.1 M ascorbic acid aqueous solution was added. The mixture was left undisturbed for 60 min in a 30 ^oC water bath. The reaction solution was then centrifuged and washed with water for three times to get the final product.

1.3 Characterization

The TEM images of the sample were carried out by a JEOL JEM-1400 Plus transmission electron microscope. The SEM images of the samples were taken by S-4800 (HITACHI, Japan) scanning electron microscopy. X-Ray diffraction patterns were measured at 25 ^oC using Rigaku. Datas were collected on Rigaku D/max 2500

instrument equipped with a Cu filter under the following conditions: scan speed, 2°/min; Cu Kα radiation, λ = 1.5405 Å. Hitachi U-3010 spectrophotometer was used to record the UV-Vis spectra of the various samples. The zeta potential was recorded by a dynamic light scattering technique (Zetasizer Nano, Malvern).

1.4 Peroxidase catalytic activity evaluation of Cu_2O

To evaluate the catalytic activity of Cu_2O hollow nanocubes, the oxidation of TMB was performed. Typically, Cu_2O (5 mg/mL, 10 μL) was dispersed into acetate buffer (0.2 M, pH = 4.5, 2.0 mL). Then TMB (0.05 M dissolved in DMSO, 10 μL) and H_2O_2 (30 wt%, 10 μL) were added into the above solution. The mixture was incubated at room temperature for 30 min, and then the absorbance data was collected using UV-vis absorption spectroscopy. The peroxidase catalytic activity of Cu_2O in different pH media was determined using the above procedure except of changing the buffer solution.

1.5 The 6-mer ssDNA sequences "turn on" Cu_2O catalytic activity at neutral pH

Cu_2O (5 mg/mL, 10 μL) and 5 μM 6-mer ssDNA sequences (A6, T6, C6, G6) were dispersed into the mixture of 50 μL pH7.4 PBS buffer and 350 μL deionized water, which was then incubated for 10 min. Then TMB (0.05 M dissolved in DMSO, 10 μL) and H_2O_2 (30 wt%, 5 μL) were added into the above solution. The mixture was reacted at room temperature for 30 min, and then the absorbance data was collected using UV-vis absorption spectroscopy.

1.6 Guanosine-rich aptamers "turn on" Cu_2O catalytic activity at neutral pH

The procedure was similar to 6-mer ssDNA sequences (A6, T6, C6, G6) except for changing the ssDNA sequences to Guanosine-rich aptamers (including IBA, KBA, HBA and TBA) with the concentration of 5 μM.

1.7 Thrombin colorimetric sensing using Cu_2O-TBA complex

Cu_2O (5 mg/mL, 10 μL) and TBA solution (30 μM, 40 μL) were dispersed into the mixture of 50 μL pH7.4 PBS buffer and 350 μL deionized water, which was then incubated at room temperature for 10 min. Then 20 μL thrombin solution with different concentration was added into the above mixture and incubated for another 10 min. After that, TMB (0.05 M dissolved in DMSO, 10 μL) and H_2O_2 (30 wt%, 5 μL) were added

into the above solution. The mixture was reacted at room temperature for 30 min, and then the absorbance data was collected using UV-vis absorption spectroscopy.

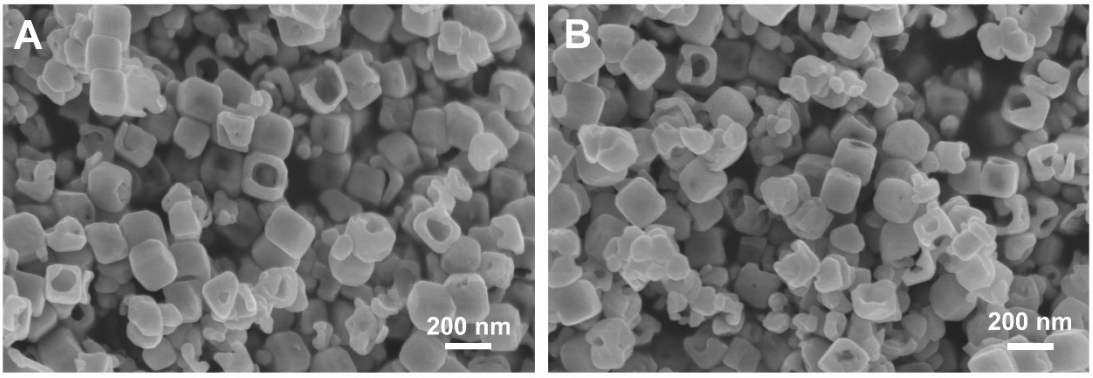

Figure S1. SEM images of Cu_2O.

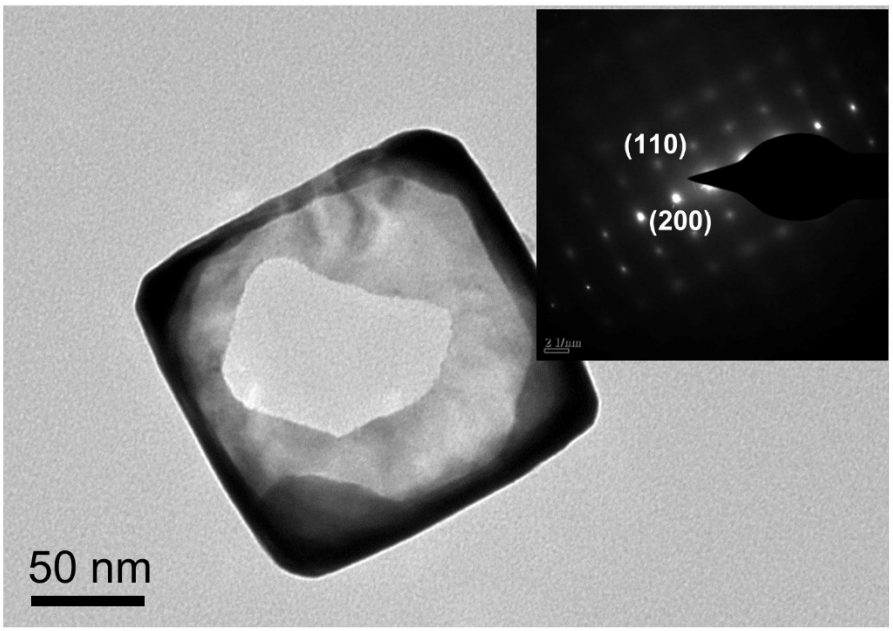

Figure S2. HRTEM image of hollow Cu_2O nanocube. Inset is the corresponding SAED pattern.

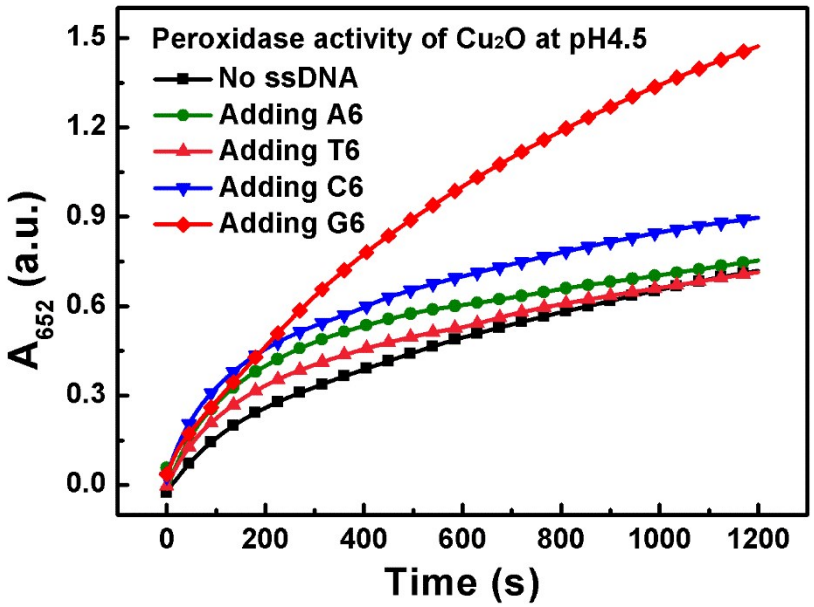

Figure S3. The effect of the addition of different ssDNA sequences on the peroxidase activity of Cu_2O at acid pH condition (pH = 4.5).

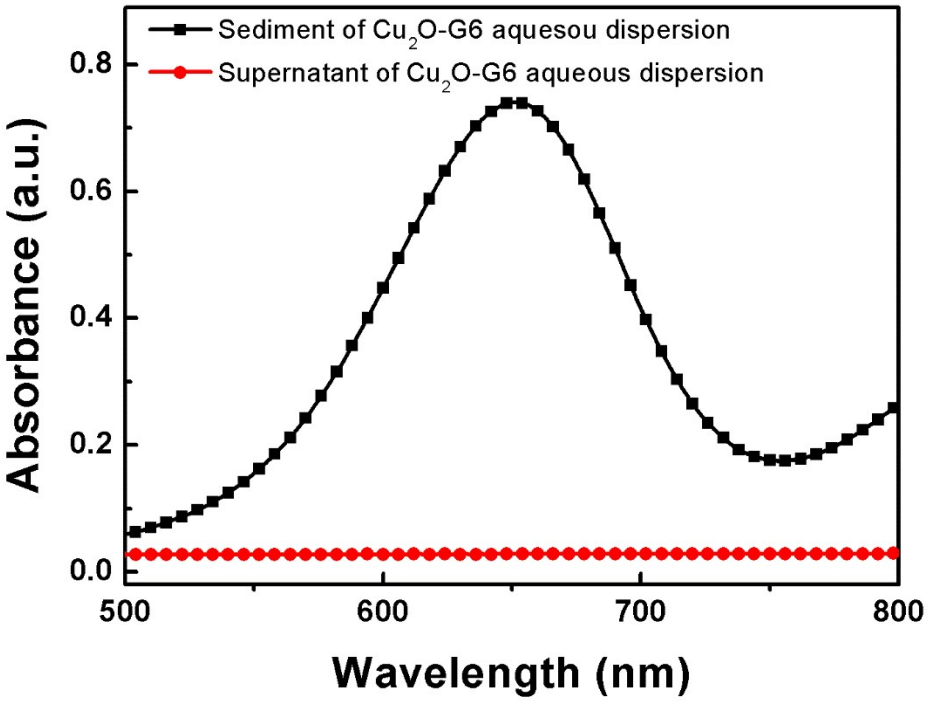

Figure S4. UV-vis absorption spectra of catalytic reaction catalyzed by the sediment and supernatant of Cu_2O-G6 aqueous dispersion using TMB as substrate in the presence of H_2O_2.

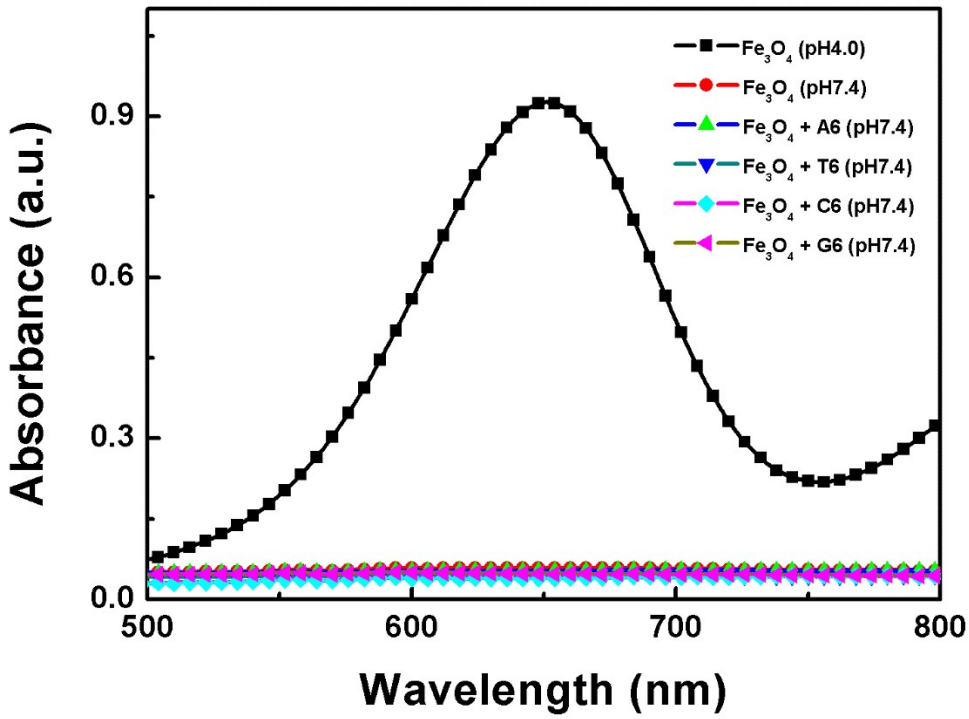

Figure S5. UV-vis absorption spectra of TMB oxidation catalyzed by Fe_3O_4 nanoparticle (at pH 4.0), Fe_3O_4 nanoparticle (at pH 7.4), Fe_3O_4 nanoparticle + A6 (at pH 7.4), Fe_3O_4 nanoparticle + T6 (at pH 7.4), Fe_3O_4 nanoparticle + C6 (at pH 7.4) and Fe_3O_4 nanoparticle + G6 (at pH 7.4) in the presence of H_2O_2.

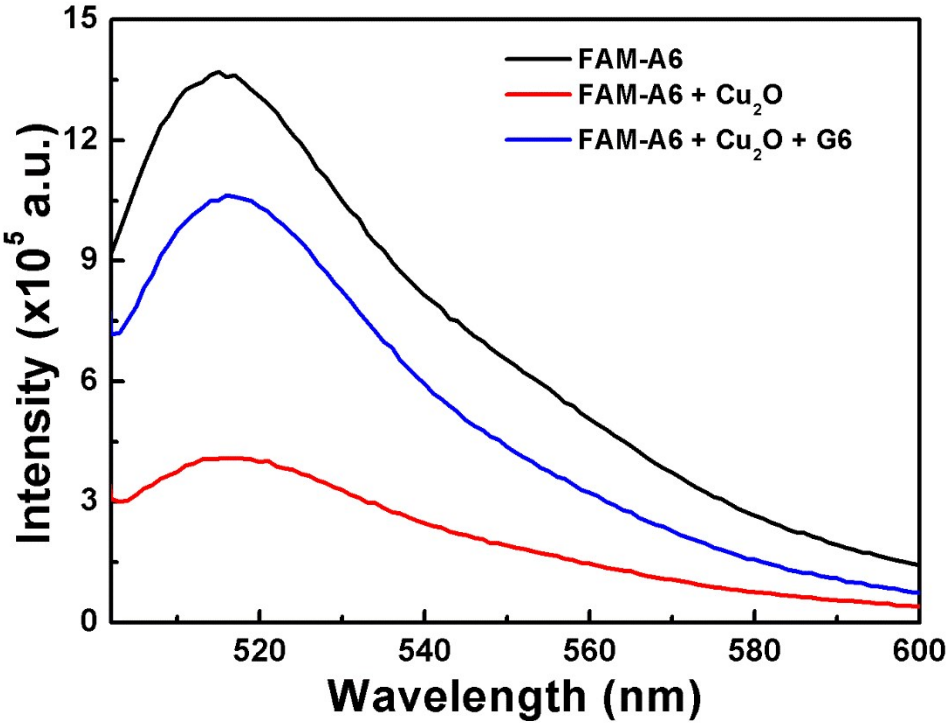

Figure S6. Fluorescence spectra of FAM-A6, FAM-A6 + Cu_2O and FAM-A6 + Cu_2O + G6.

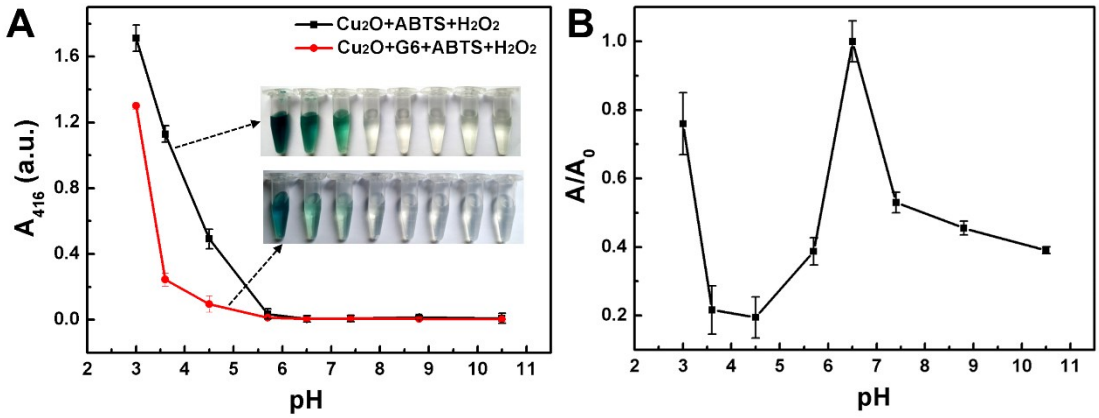

Figure S7. (A) Comparison of oxidation reaction of ABTS by Cu_2O in the absence and presence of G6 at different pH values; (B) The absorbance ratio changes of oxidation reaction of ABTS by Cu_2O after and before the addition of G6 at different pH values.

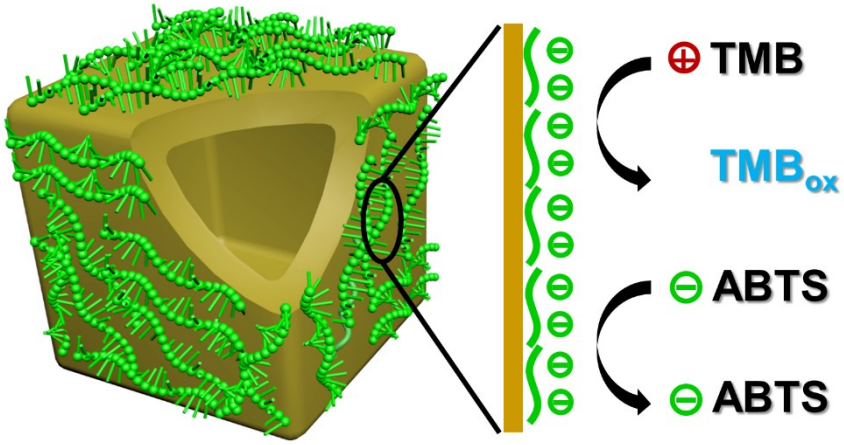

Figure S8. Illustration of the mechanism for the enhancement of peroxidase activity of the Cu_2O toward TMB.

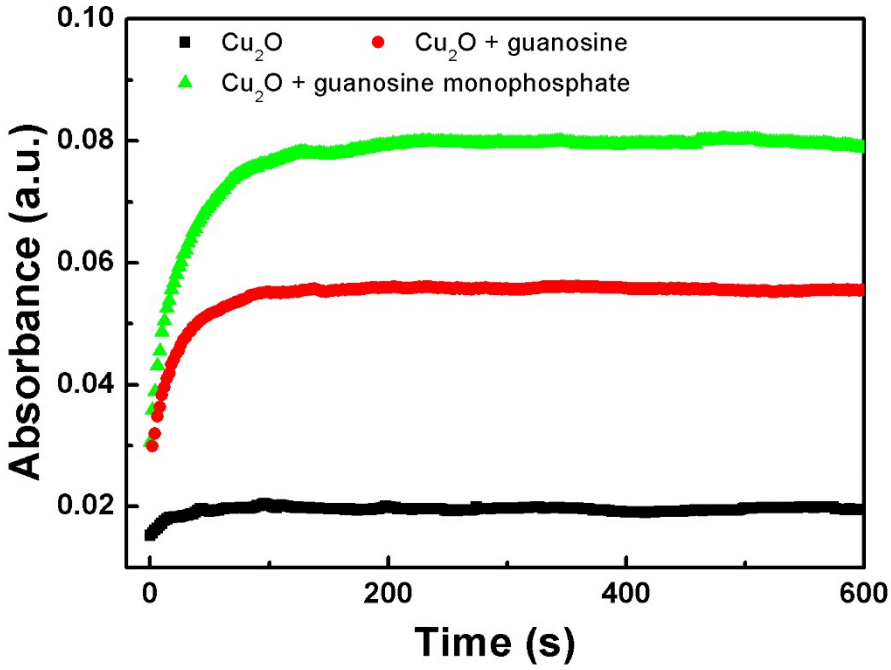

Figure S9. The enhancement of Cu_2O peroxidase activity by guanosine and guanosine monophosphate at neutral pH condition (pH=7.4).

Table S1. DNA sequences used in this study.

DNA Name	Sequence (5’ to 3’)
A6	AAAAAA
T6	TTTTTT
G6	GGGGGG
C6	CCCCCC
FAM-A6	FAM-AAAAAA
IBA	GGTGGGGGTTGGTAGGGTGTCTTC
KBA	TGGGGGTTGAGGCTAAGCCGA
HBA	TAGCGATACTGCGTGGGTTGGGGCGGGTAGGGCCA GCAGTCTCGT
TBA	GGTTGGTGTGGTTGG

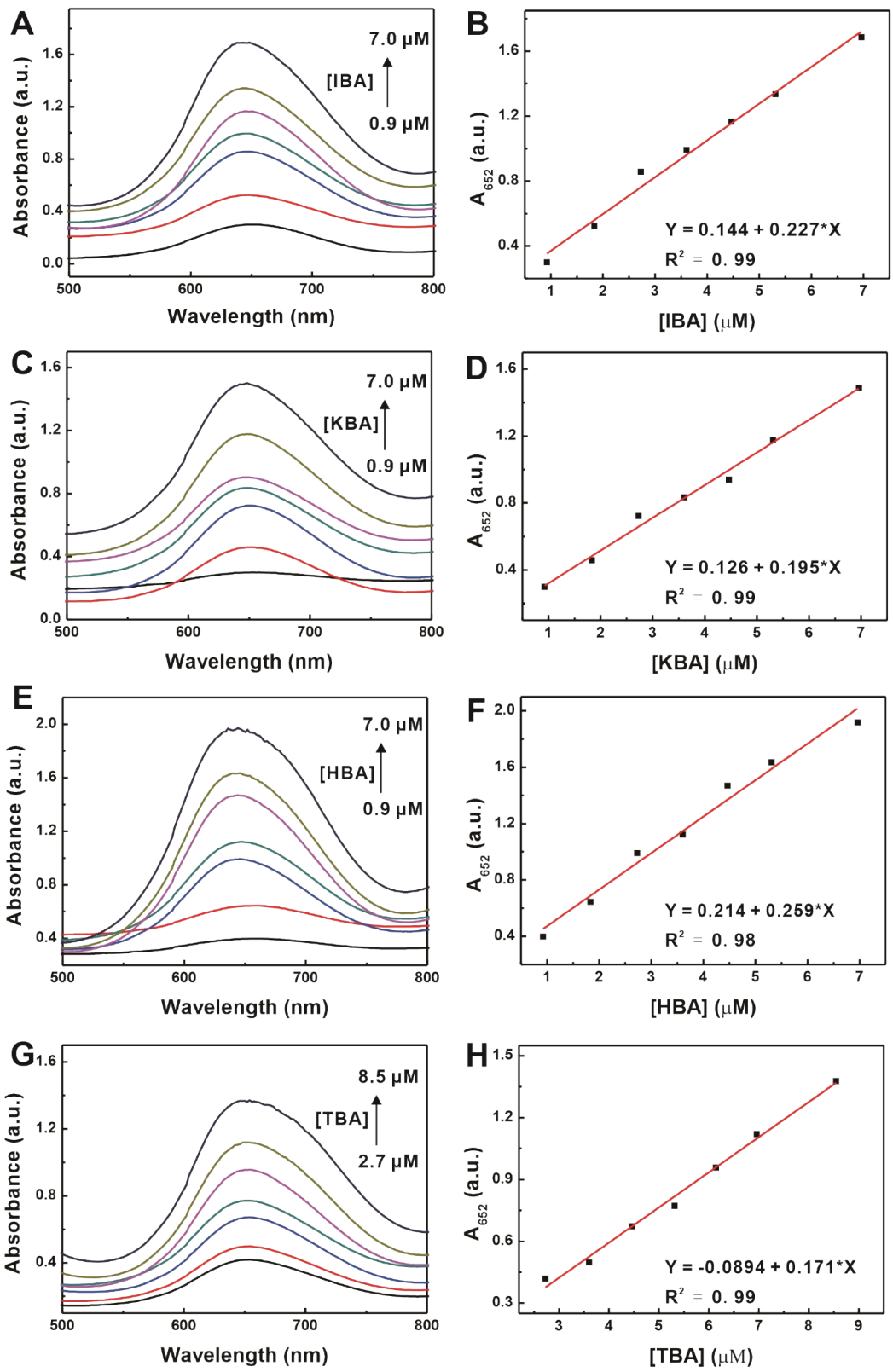

Figure S10. The enhancement of peroxidase activity of Cu_2O by G-rich aptamers with different concentrations (A) IBA; (C) KBA; (E) HBA; (G) TBA. Linear response of the Cu_2O peroxidase activity to aptamer concentration (B) IBA; (D) KBA; (F) HBA; (H) TBA.