

Supplementary Information

A Simple Lateral Flow Biosensor for the Rapid Detection of Copper (II) Ion Based on Click Chemistry

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Table S1 Sequences of the DNA used in this study

Name	Sequence (5'→3')
Azide-DNA	ATACTCCCCCAGGTGCCG
Alkyne/biotin-DNA	AGCTTCTTTCTAATACG
Control zone-DNA	CGTATTAGAAAGAAGCTCGTATTAGAAAGAAGCT
Test zone-DNA	CGGCACCTGGGGGAGTATCGGCACCTGGGGGAGTAT

Table S2 Measurements of Cu²⁺ spiked in tap water and human serum

Samples	Cu ²⁺ (μM) (n=3)		Recovery (%)
	spiked	detected	
Tap water	2	1.92 ± 0.09	91.3%
	20	22.8 ± 0.79	109.3%
	200	190.4 ± 1.92	95.2%
Human serum	2	1.91 ± 0.12	95.5%
	20	19.2 ± 1.09	96%
	200	208.4 ± 1.92	104.2%

Table S3 Assessment of repeatability and reproducibility of the biosensor

researcher	optical intensity					CV
1	924	830	832	917	869	4.06%
	895	827	842	859	912	
	920	816	832	891	870	
2	830	920	811	870	845	4.38%
	823	831	865	841	851	
	921	890	845	835	824	
CV	4.23%					

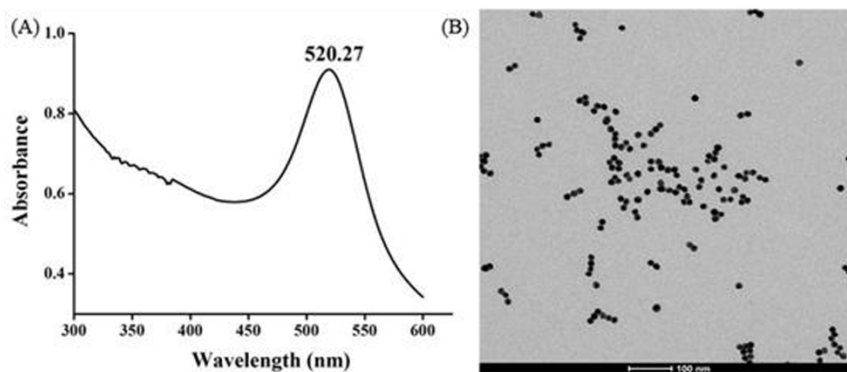


Fig. S1 (A) The UV absorption spectrum and (B) TEM photographs of AuNPs used in the experiment. Scale bars are 100 nm.

	1	2	3	4	5	6	7	8	9
Azide	2	2	2	2	2	2	2	2	2
DNA	uM	uM	uM	uM	uM	uM	uM	uM	uM
Cu ²⁺	0	200	200	200	200	200	0	0	0
		uM	uM	uM	uM	uM			
VC	0	500	500	500	500	500	0	0	0
		uM	uM	uM	uM	uM			
H ₂ O ₂	0	0	200	500	1	2	500	1	2
			uM	uM	mM	mM	uM	mM	mM

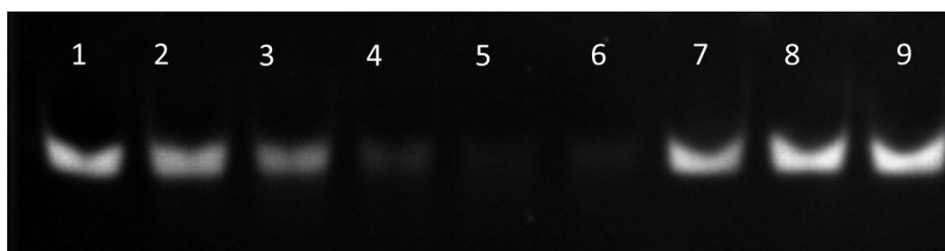


Fig. S2 PAGE of the azide-DNA under different concentrations of Cu²⁺, sodium ascorbate and H₂O₂ and ligation product. We designed an experiment to measure the DNA scission. We treated the azide-DNA with different concentrations of Cu²⁺, sodium ascorbate and H₂O₂ and ran on PAGE. When only 200μM Cu²⁺/500μM sodium ascorbate were added to 2μM azide-DNA, no extensive DNA degradation occurred, the same thing happen when only H₂O₂ was added even at a high concentration of 2 mM. But when 200μM Cu²⁺/500μM sodium ascorbate was mixed with different concentrations of H₂O₂, DNA degradation occurred, and with the increase of H₂O₂, DNA degradation increased. In our method, no extra H₂O₂ was added, so there was not so much H₂O₂ produced by the reaction of ascorbate acid and Cu²⁺. Therefore no extensive DNA degradation occurred.

	1	2	3
Alkyne	2	2	2
DNA	uM	uM	uM
Cu ²⁺	0	200	200
		uM	uM
VC	0	500	500
		uM	uM
H ₂ O ₂	0	0	500
			uM

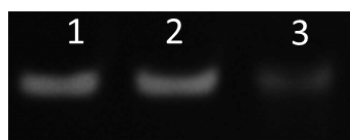


Fig. S3 PAGE of the alkyne-DNA under different concentrations of Cu²⁺, sodium ascorbate and H₂O₂.