Supplementary Material

A sensitive and regenerative electrochemical immunosensor for quantitative detection of *Escherichia coli* O157:H7 based on stable polyaniline coated screen-printed carbon electrode and rGO-NR-Au@Pt

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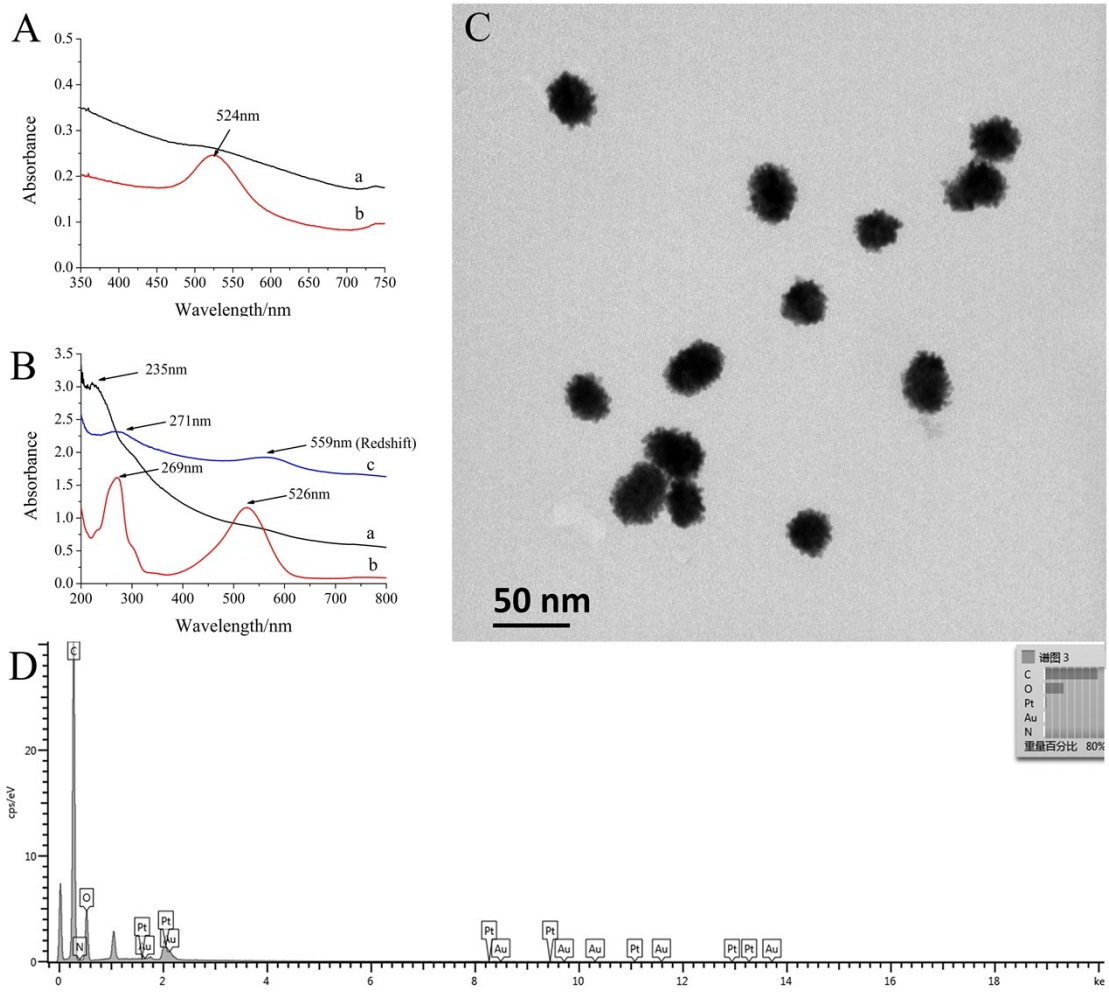


Fig. S1 (A) UV-vis spectra of (a) Au@Pt nanoparticles and (b) AuNPs nanoparticles; (B) UV-vis spectra of (a) GO, (b) NR and (c) rGO-NR; (C) TEM image of Au@Pt nanoparticles; (D) EDX image of rGO-NR-Au@Pt.

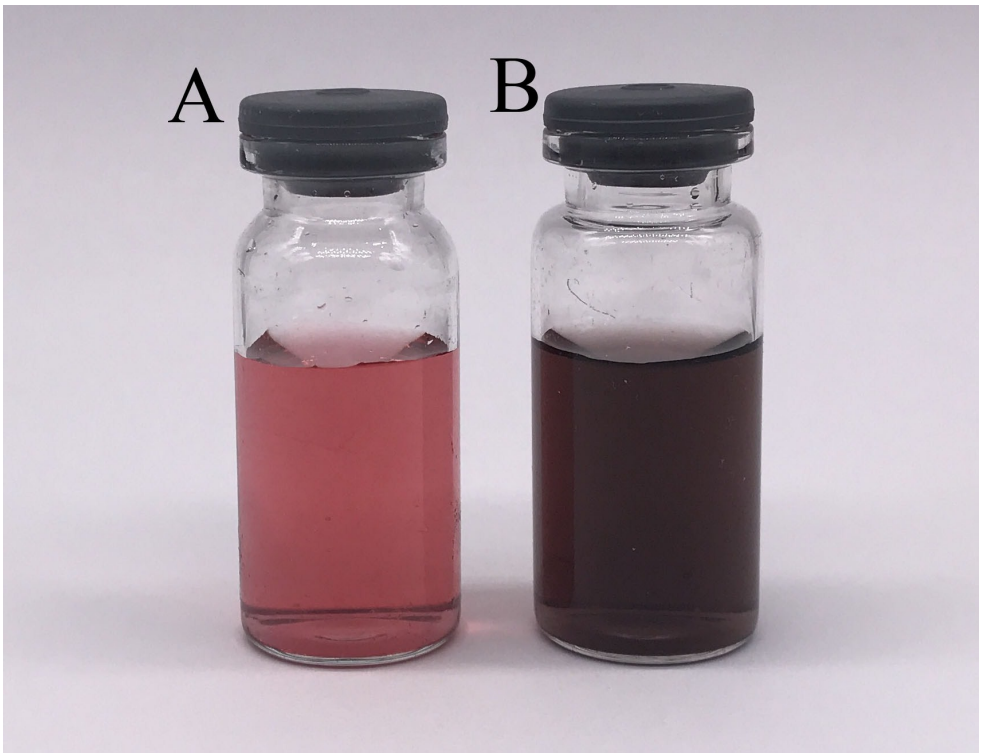


Fig. S2 The color comparison chart of the (A) AuNPs and (B) Au@Pt nanoparticles.

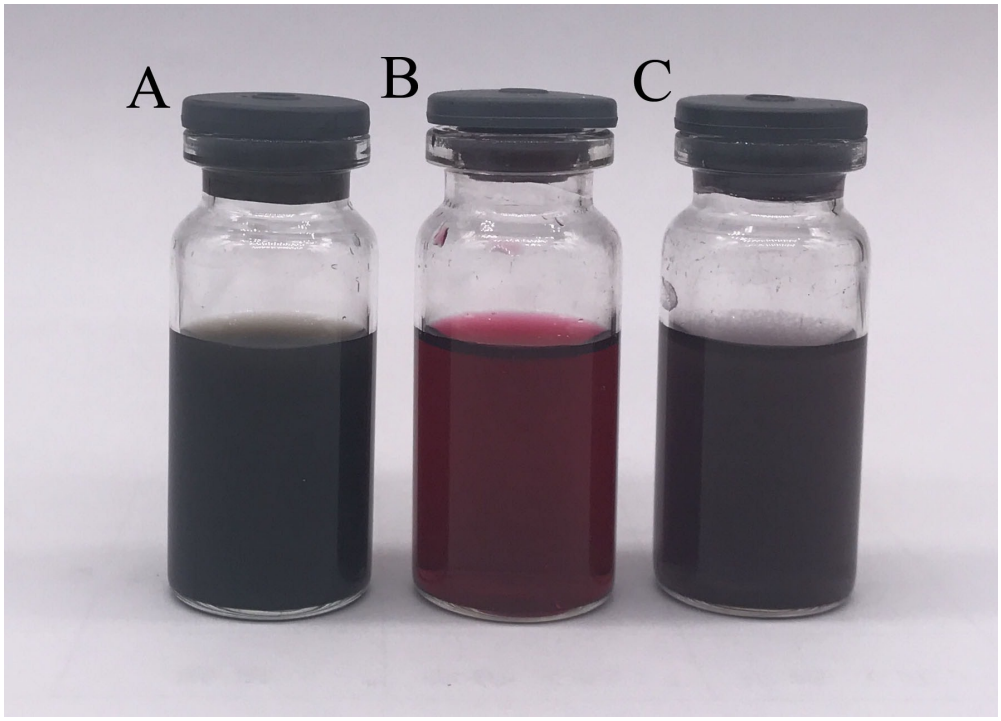


Fig. S3 The color comparison chart of the (A) GO, (B) NR and (C) rGO-NR.

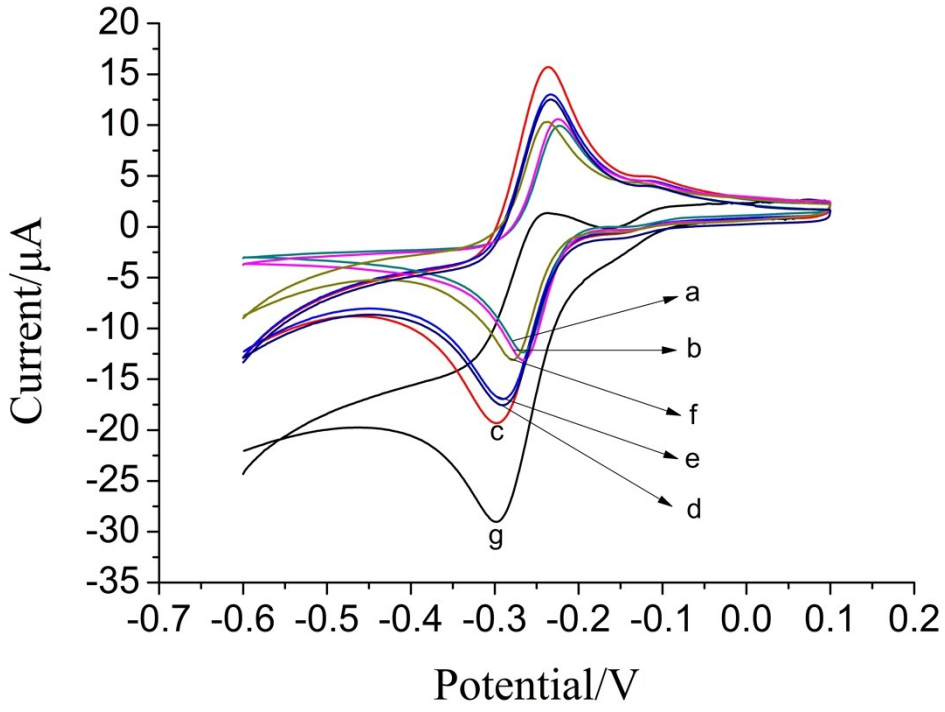


Fig. S4 CVs of (a) Bare SPCE, (b) SPCE-PANI, (c) SPCE-PANI-AuNPs, (d) SPCE-PANI-AuNPs-Ab_1, (e) SPCE-PANI-AuNPs-Ab_1 -BSA, (f) SPCE-PANI-AuNPs-Ab_1 -BSA -*E. coli* O157:H7 and (g) SPCE-PANI-AuNPs-Ab_1 -BSA -*E. coli* O157:H7- rGO-NR-Au@Pt -Ab_2 recorded in HAc-NaAc (pH=5.5, 0.1 M) containing 1.0 mM TH and 5.0 mM H_2O_2.

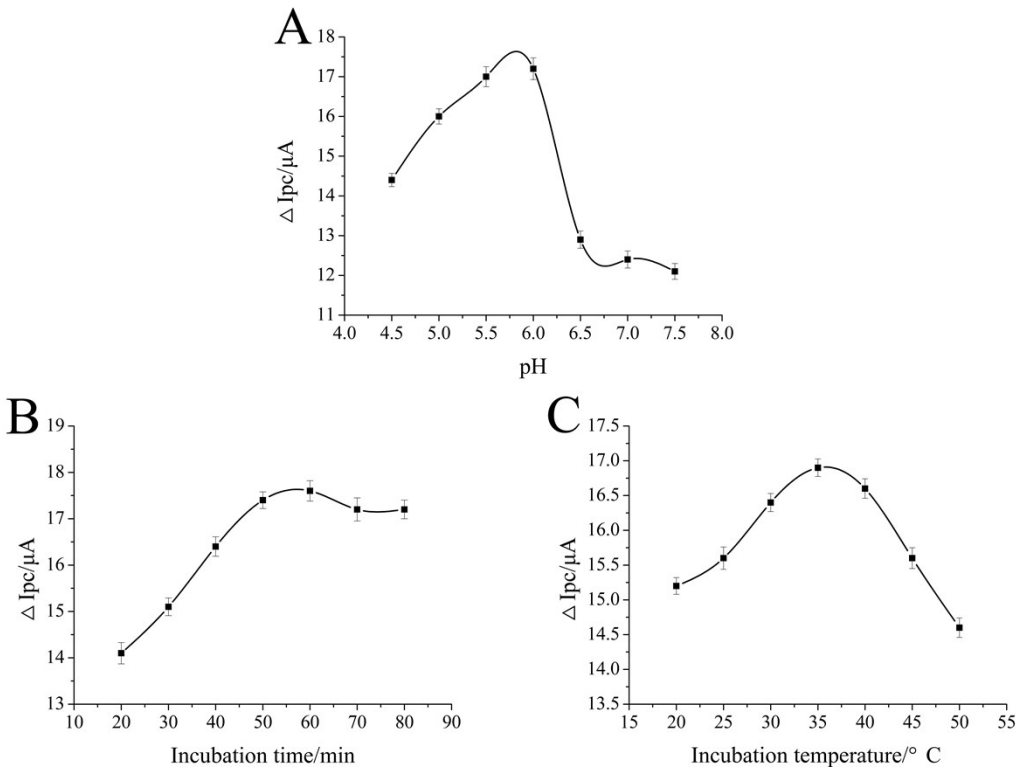
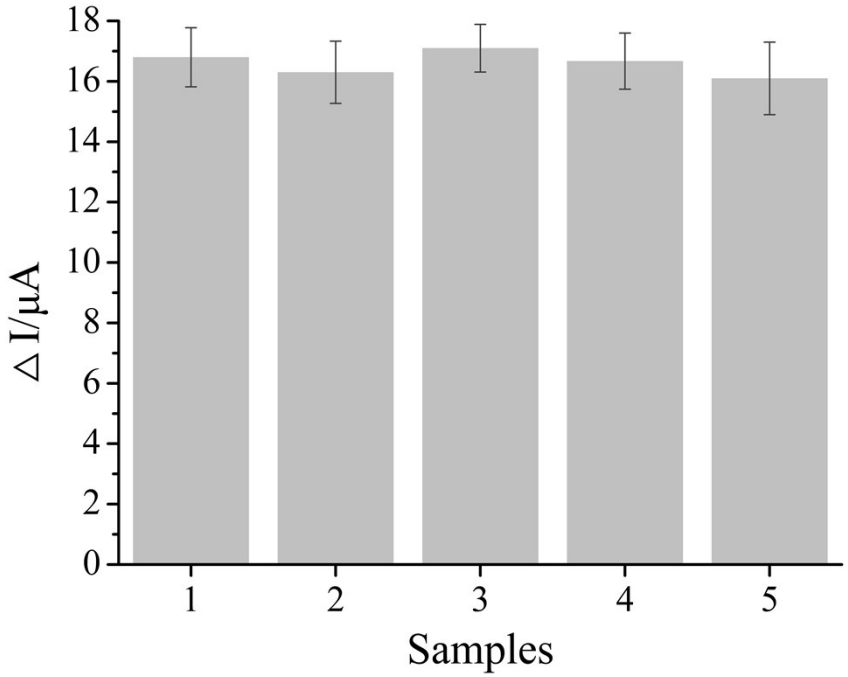


Fig. S5 Optimization of experimental parameters: (A) Influence of working buffer pH; (B) Influence of incubation time between *E. coli* O157:H7 and rGO-NR-Au@Pt-Ab_2-BSA; (C) Influence of incubation temperature between *E. coli* O157:H7 and rGO-NR-Au@Pt-Ab_2-BSA.



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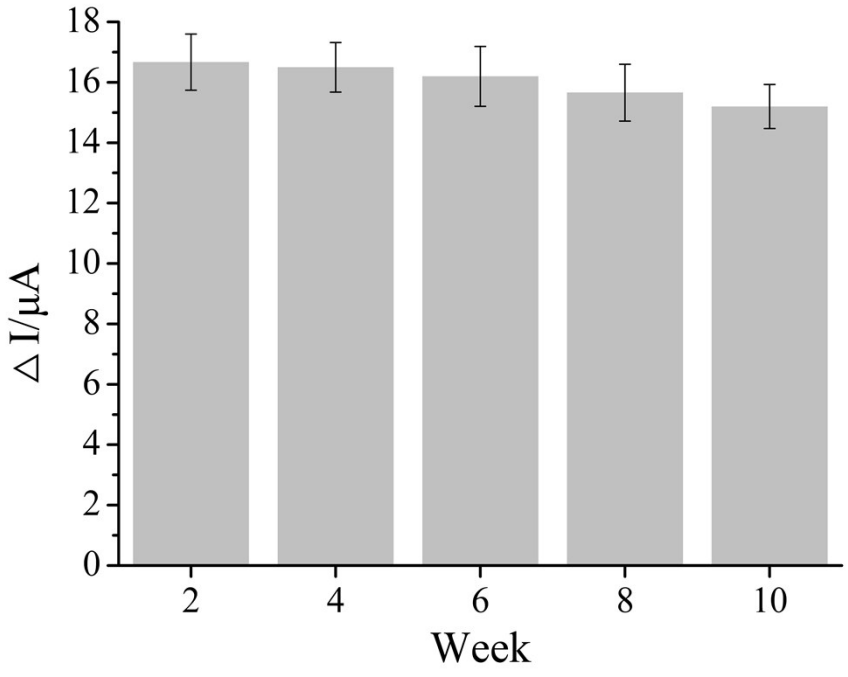
36 **Fig. S6** The reproducibility study of the immunosensor.



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38 **Fig.S7** The stability study of the immunosensor.

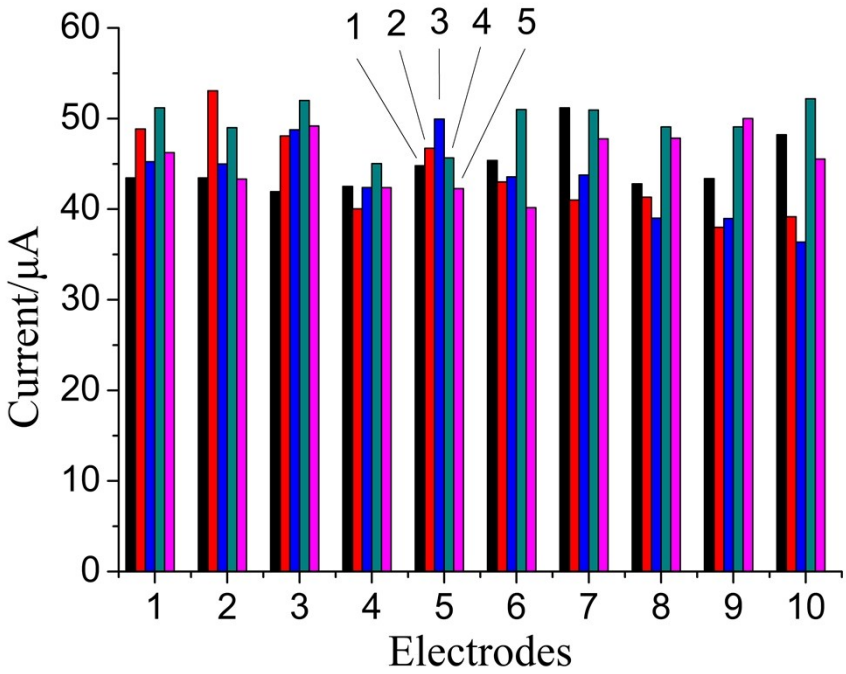

Fig. S8 The regeneration data of the immunosensor. Ten electrodes, each electrode regenerated four times, tested for the same concentration of *Escherichia coli* O157:H7.

Table. S1

Comparison of the analytical parameters for *E. coli* O157:H7 detection obtained in this study and reported by previous studies.

Methods	Linearity range (CFU·mL^{-1})	LOD (CFU·mL^{-1})	Ref.
EIS	1.0×10^4 -1.0×10^7	1.0×10^4	[31]
SPR	3.0×10^4 -3.0×10^8	3.0×10^4	[32]
CL	3.0×10^4 -3.0×10^6	3000	[33]
Fluorescent	5.0×10^3-5.0×10^6	5000	[34]
DPV	4.0×10^4- 4.0×10^9	7.98×10^3	our previous article
CV	8.9×10^3- 8.9×10^9	2.84×10^3	this work

LOD: limit of detection. **EIS**: Electrochemical impedance spectroscopy. **SPR**: Surface plasmon resonance. **CL**: Chemiluminescence. **DPV**: differential pulse voltammetry

Table. S2

Accuracy experimental results of the immunosensor in milk samples.

Sample (milk)	1	2	3	4	5	6	7	8	9	10	11	12
Actual results	-	+	-	-	+	+	+	-	+	+	-	-
Test results	-	+	-	+	+	-	+	-	+	+	-	-

(In actual results: '+' means add a certain concentration of *E. coli* O157:H7, '-' means negative samples; In test results: '+' means $\Delta I > 9.27$ μA, '-' means $\Delta I < 9.27$ μA.)

Table. S3

Accuracy experimental results of the immunosensor in pork samples.

Sample (pork)	1	2	3	4	5	6	7	8	9	10	11	12
Actual results	+	+	-	+	-	-	-	+	+	-	+	-
Test results	+	+	-	+	-	-	+	+	+	-	+	-

(In actual results: '+' means add a certain concentration of *E. coli* O157:H7, '-' means negative samples; In test results: '+' means $\Delta I > 9.27$ μA, '-' means $\Delta I < 9.27$ μA.)

Table. S4

Detection results and recoveries of the immunosensor in milk and pork samples.

Sample	Spiked (CFU/mL)	ΔI (μA)	Found (CFU/mL)	Recovery (%)	RSD (%, n=3)
Milk1	8.9×10^4	11.215	8.1×10^4	91.01	4.7
Milk2	8.9×10^4	11.130	7.0×10^4	78.65	2.5
Milk3	8.9×10^6	13.834	7.4×10^6	83.15	5.1
Milk4	8.9×10^6	13.818	7.2×10^6	80.90	3.9
Milk5	8.9×10^8	16.513	7.5×10^8	84.27	4.1
Milk6	8.9×10^8	16.558	8.1×10^8	91.01	3.4
Pork1	8.9×10^4	11.307	9.5×10^4	106.74	4.3
Pork2	8.9×10^4	11.222	8.2×10^4	92.13	3.7
Pork3	8.9×10^6	13.921	8.6×10^6	96.63	4.6
Pork4	8.9×10^6	13.872	7.9×10^6	88.76	2.8
Pork5	8.9×10^8	16.632	9.2×10^8	103.37	3.6
Pork6	8.9×10^8	16.572	8.3×10^8	93.26	4.2