SUPPLEMENTARY INFORMATION

Hierarchical Cystine Flower Based Electrochemical Genosensor for Detection of *Escherichia coli* O157:H7

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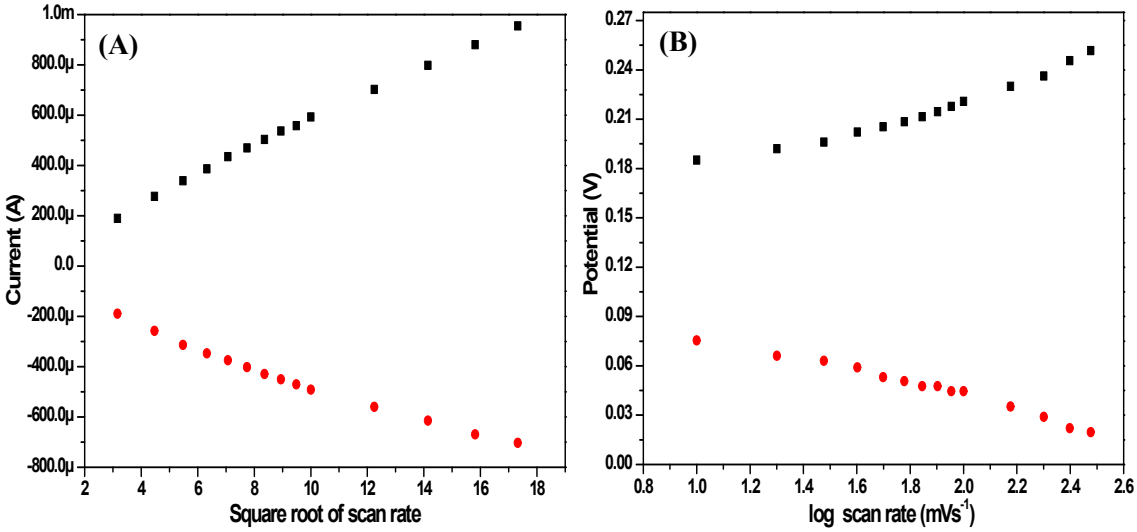
Figure S1: Cyclic voltammogram peak current analysis of of pDNA/CysFl/Au bioelectrode as a function of scan rate (10-300 mV/s). **(A)** Variation of current with square root of scan rate and **(B)** variation of potential with log of scan rate.



3.6.2. Estimation of the association constant between cDNA and pDNA/CysNf/Au bioelectrode.

The association constant (K_a) for binding interaction was determined using the Langmuir isotherm approach in which the isotherm assumes equal binding energy for all binding sites. For K_a estimation, the change in Rct was related to the binding of cDNA with immobilized pDNA and is represented by the following equation:

$$M = 1- R_{ct}(pDNA)/R_{ct}(cDNA) \quad (1)$$

where M, is the number of occupied binding sites and R_{ct}(pDNA), R_{ct}(cDNA) represents the charge transfer resistance before and after hybridization with cDNA, respectively. In the Langmuir isotherm, M can be related to association constant using the following equation:

$$M = K_aC /1+K_aC \quad (2)$$

$$K_aC = M /1\text{-}M \quad (3)$$

where K_a is the association constant and C is the concentration of molecules in the solution. From eqs 2 and 3 above

$$K_aC = R_{ct}(cDNA) – R_{ct}(pDNA) / R_{ct}(pDNA) \quad (4)$$

$$K_aC = \Delta R_{ct} / R_{ct}(pDNA) \quad (5)$$

Using eq 5, the curve was plotted between $\Delta R_{ct}/R_{ct}(pDNA)$ and the concentration of cDNA (Fig. S2) and it was revealed that $\Delta R_{ct}/R_{ct}(pDNA)$ varies linearly with the concentration and follows the linear equation

$$\Delta R_{ct}/R_{ct}(pDNA) = 3.5852 + 0.2432 \log cDNA \quad (6)$$

with a correlation coefficient of 0.998. K_a was estimated from the slope of the regression equation and found to be 0.243 M^{-1}.

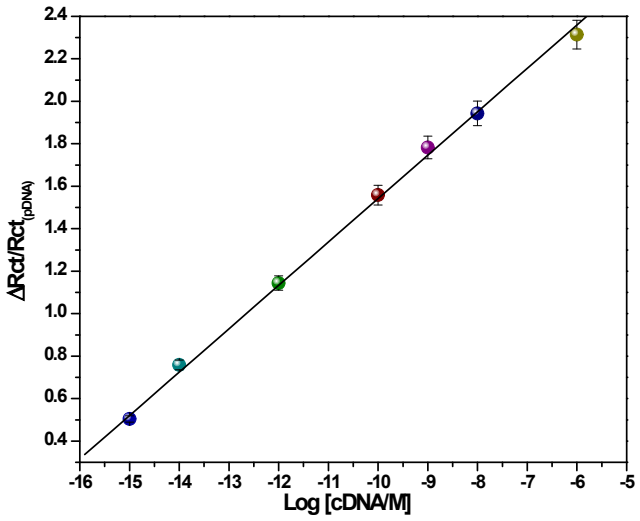


Figure S2. Linearity plot between $\Delta R_{ct}/R_{ct(pDNA)}$ as a function of cDNA concentration (10^{-6}-10^{-15} M) in 5 mM $[Fe(CN)_6]^{3-/4-}$ PBS solution at pH 7 for measurement of K_a.

Table S1

S. No.	Bare gold (°)	CysFl/Au (°)	pDNA/CysFl/Au (°)					
			2h	4h	6h	8h	12h	18h
1	97.96	54.95	42.46	31.21	29.65	29.13	28.98	28.42
2	97.92	55.03	42.35	30.98	29.02	29.23	29.08	28.93
3	97.83	54.69	41.98	30.95	29.38	28.82	29.12	28.63
4	97.90	54.83	42.42	30.89	29.68	29.20	29.18	29.22

Table S1: Variation in the contact angle of Au, CysFl/Au, and pDNA/CysFl/Au ODT/Au as a function of time.

Table S2

Sl. No	Electrode	Solution resistance (R_s, Ω)	Charge transfer resistance (R_{ct}, Ω)
1	CysFl/Au	84.2	250.4
2	EDC/NHS activated CysFl/Au	24.0	16.7
3	pDNA/CysFl/Au	24.2	47.5
4	Complementary DNA on pDNA/CysFl/Au	74.0	149.3

Table S2: The change in R_{ct} value for CysFl/Au, EDC/NHS activated CysFl/Au, pDNA/CysFl/Au and complementary DNA on pDNA/CysFl/Au as a function of time.

Table S3

No. of cycle	Before hybridization (R_{ct}, Ω)	After hybridization (R_{ct}, Ω)	% change in hybridization efficiency
1	47.28	148.23	0.76
2	47.23	146.07	2.21
3	47.02	143.04	4.24
4	46.88	139.89	6.35
5	46.54	136.26	8.78
6	46.02	133.45	10.37

Table S3: Comparison of the change in hybridization efficiency for pDNA/CysFl/Au bioelectrode with repeated denaturation and rehybridization process.