Supplementary Information

Electrochemically-Driven and Dynamic Enhancement of Drug Metabolism *via* Cytochrome P450 Microsomes on Colloidal Gold/Graphene Nanocomposites

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Figure S1. The typical TEM images of Au NPs.

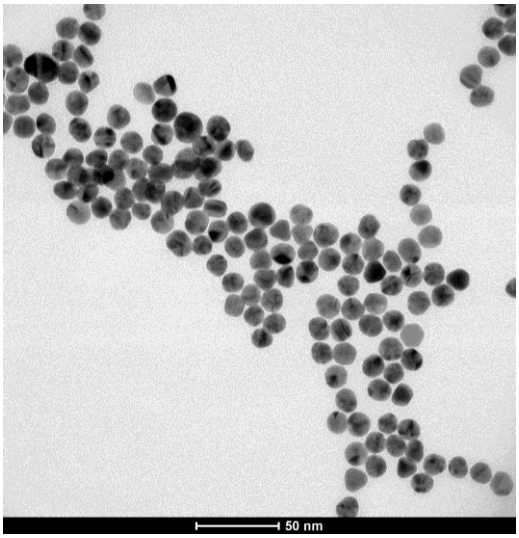


Figure S2. Optical photographs of (A) PDDA/G and (B) unmodified graphene.

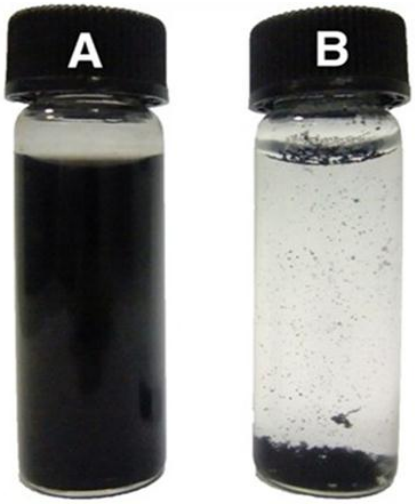


Figure S3. Raman spectra of (a) graphene oxide and (b) PDDA/G.

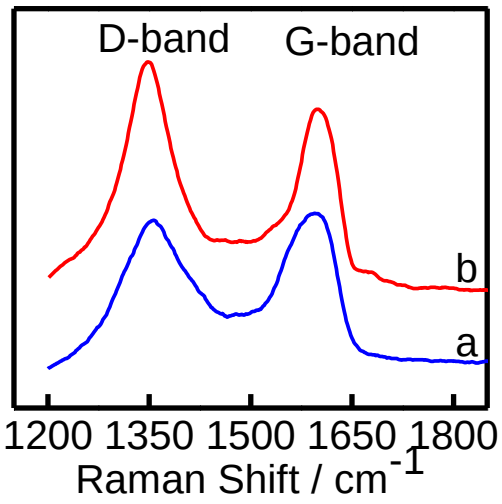
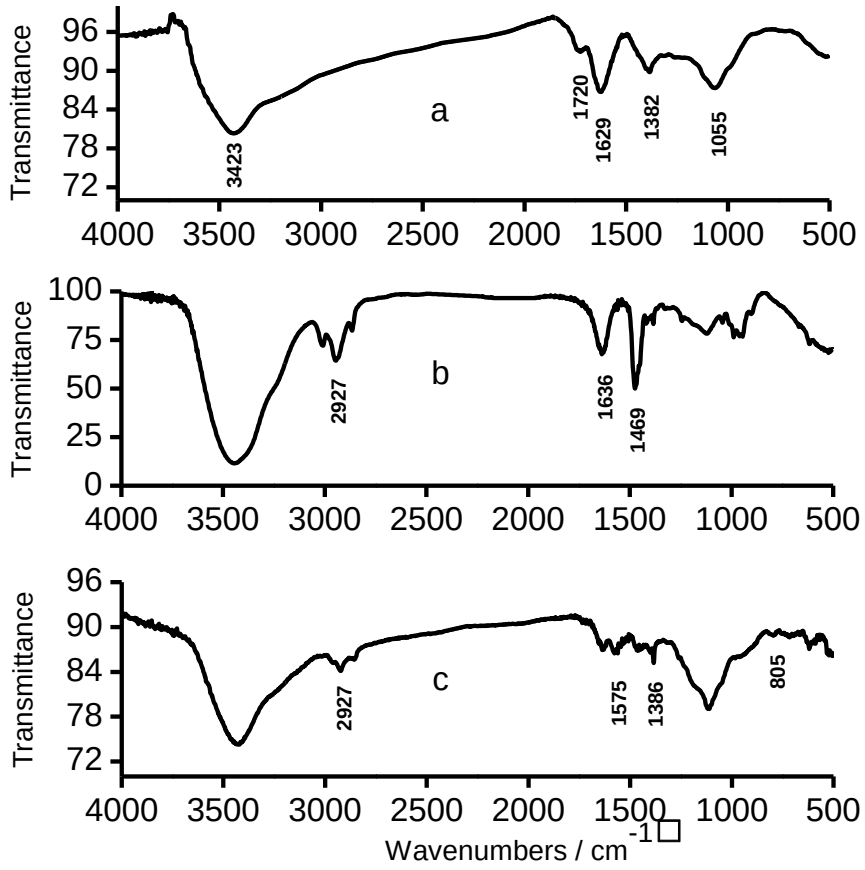


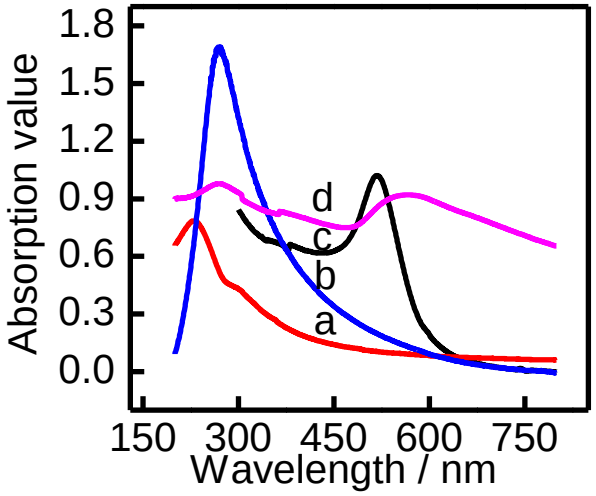
Figure S4. FTIR spectra of (a) GO, (b) PDDA, (c) PDDA/G.



The IR spectrum of the graphene oxide (Fig. S4a) showed a C-O stretch peak at 1055 cm^{-1}, a strong C=O stretch at 1720 cm^{-1}, as well as O-H stretch at 1382 cm^{-1} and 3423 cm^{-1}. These peaks indicated the presence of oxygen functional groups on the surface of GO.[1] The peak at 1629 cm^{-1} could be attributed to the stretching deformation vibration of intercalated water and the skeletal vibrations of unoxidized graphitic domains.[2] Compared with GO, the spectrum of PDDA/G (Fig. S4c) showed an obvious skeletal vibration adsorption band of graphene at 1575 cm^{-1}, whereas most of other adsorption bands of oxygen functionalities disappeared (C=O, epoxy groups etc). The disappearance of these bands suggested considerable deoxygenation by the

chemical reduction process once again. Besides this, two new peaks at 1385 cm^{-1} (N-O) and 805 cm^{-1} (C-N) appeared which indicated that nitroso groups were produced in the presence of PDDA. In the case of PDDA and PDDA/G, both spectra showed the characteristic absorption bands of PDDA, such as the CH_2 asymmetrical and symmetrical stretching frequencies (2927 cm^{-1}) and C=C stretching vibrations (1639 cm^{-1}, 1469 cm^{-1}).[3]

Figure S5. UV-*vis* absorption spectra of (a) GO, (b) PDDA/G, (c) Au-NPs and (d) Au/PDDA/G.



The reduction process and functionalized process was then characterizated by UV-vis absorption spectra as shown in Fig S3. The maximum absorption transferred from 230 nm of GO (curve a) to 268 nm of PDDA-G (curve b). This change indicated that the electronic conjugation in the graphene sheets was restored after the reduction.[4] The UV-vis absorption spectra of citrate-coated Au NPs was centered at 518 nm (curve c), which was consistent with well-dispersed, spherical Au NPs in aqueous media. After being modified with Au nanoparticles, the Au/PDDA/G nanocomposites produced a clearly absorbance at 268 nm, corresponded to the overlap of absorbance bands of PDDA/G, and a broaden absorbance band at 559 nm, for the absorbance of Au nanoparticles itself (curve d).

Figure S6. Measured average zeta potential of GO, PDDA/G, Au/PDDA/G, PDDA/Au/PDDA/G.

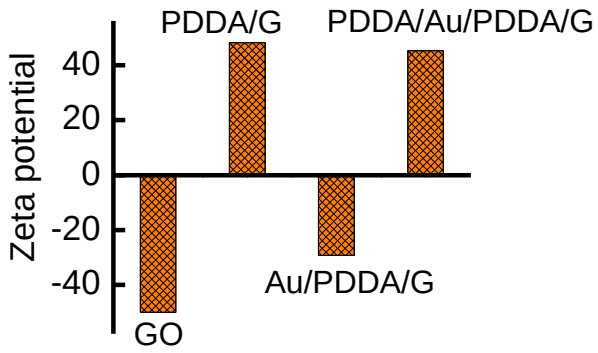


Figure S7. CV curves of CYP3A4/CPR-microsomes/PDDA/Au/PDDA/G modified GCE at scan rate of 100, 200, 300, 400, 500 mV s^{-1} (from inner to outer curve) in anaerobic 0.1 M PBS, pH 7.4. Inset: plots of peak currents vs. scan rates.

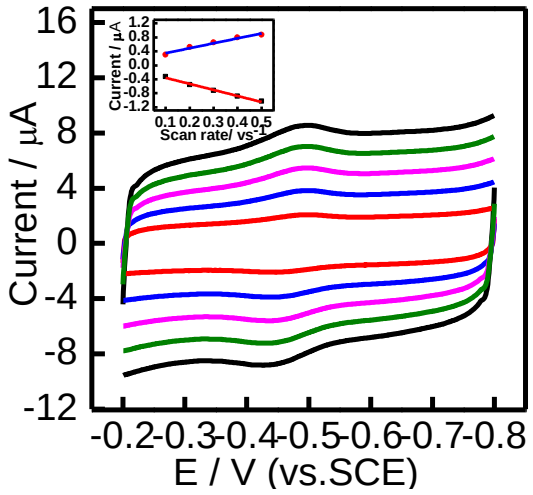


Figure S8. CV curves obtained at (a) PDDA/G and (b) CYP3A4/CPR-microsomes/PDDA/G modified GCE at a scan rate of 100 mV s^{-1} in anaerobic 0.1 M PBS, pH 7.4.

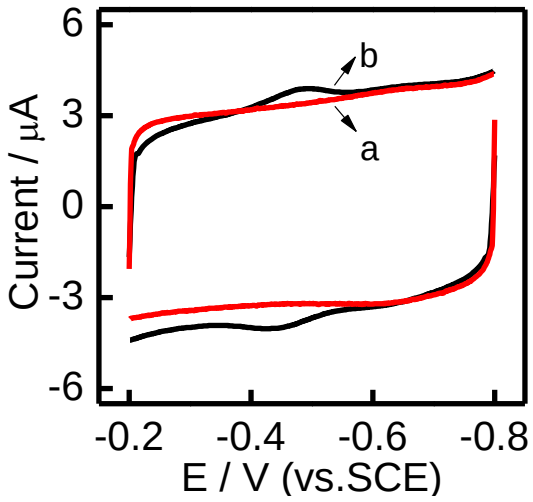
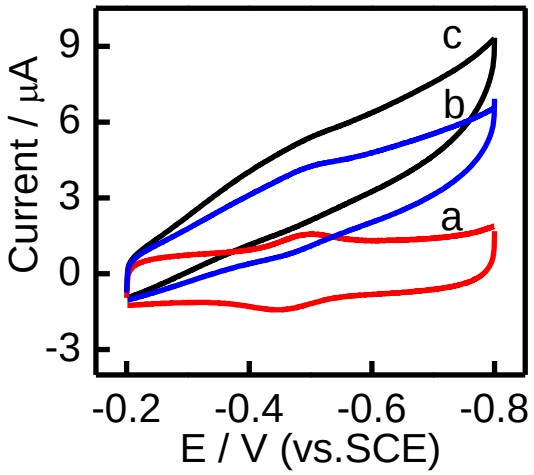
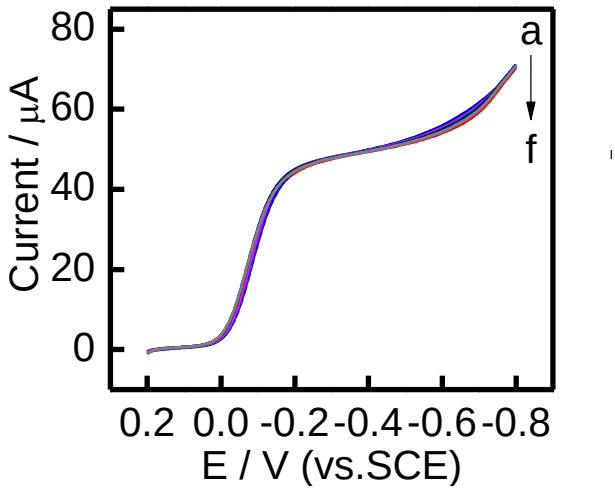


Figure S9. CV curves of CYP3A4/CPR-microsomes/PDDA/Au/PDDA/G modified GCE in 0.1 M PBS, pH 7.4 at 100 mV s^{-1}. (a) Nitrogen saturated, (b) air saturated, and (c) with the addition of 46μM nifedipine to air saturated buffer solution.



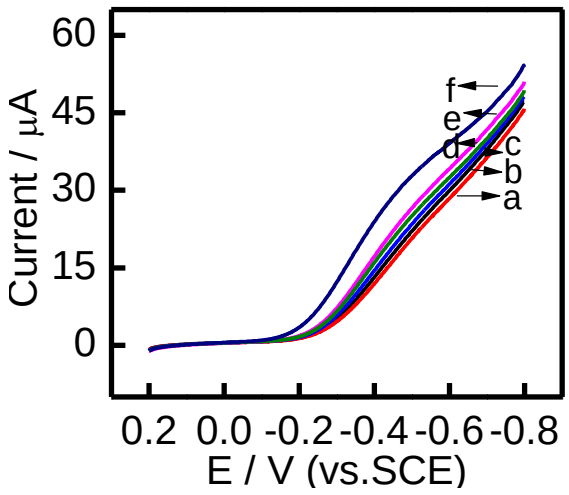
In the presence of oxygen, greatly increased reduction peaks (at 100 mV s^{-1}) and almost no oxidation peaks were observed (supporting information Fig S9b), which suggested that a typical electrocatalytic oxygen reduction process based on fast binding of oxygen to the reduced ferrous cytochrome P450. This was in good agreement with the previous reports on electrocatalytic oxygen reduction of cytochrome P450. When nifedipine was added to the air-saturated buffer solution, an increase of the reduction current at CYP3A4/CPR-microsomes/PDDA/Au/PDDA/G/GCE was observed (supporting information Fig S9c).

Figure S10. PDDA/Au/PDDA/G films in electrochemical biocatalysis. Influence of increasing nifedipine concentration on rotating disk voltammograms (1000 rpm) in aerobic 0.1 M PBS, pH 7.4.



In the absence of the CYP3A4/CPR-microsomes, no similar cathodic peak corresponding to the oxidation of nifedipine was observed from the PDDA/Au/PDDA/G/GCE.

Figure S11. CYP3A4/CPR-microsomes/PDDA/Au/PDDA/G films in electrochemical biocatalysis. Influence of successive addition of different reagents of O_2 (a), enthonal (b), PBS (c), distilled water (d), tolbutamide (e), nifedipine (f) on rotating disk voltammograms (1000 rpm) in aerobic 0.1 M PBS, pH 7.4.



Only slight response signals on CYP3A4/CPR-microsomes/PDDA/Au/PDDA/G/GCE were observed when an equivalent aliquot of other reagents (distilled water, enthonal, PBS, tolbutamide) were injected into the solution. This behavior was characteristic of the electrochemical response of some heme proteins immobilized on an electrode and corresponded to the reduction of dissolved oxygen.[5,6,7] When nifedipine was added to PBS, an obvious catalytic peak was observed.

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