Supplementary Information

A Comparative Evaluation on Activity Modulation of Flavo and non-Flavo EnzymesInduced by Graphene Oxide

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Methods

Synthesis of Graphene Oxide

Graphitic oxide was prepared by the oxidative treatment of natural graphite using Hummer's method.¹ In brief, 0.5 g of graphite flakes, 0.25 g of sodium nitrate were mixed in a beaker which was kept in an ice bath and then sulfuric acid (12 mL) was added with vigorous stirring. Maintaining vigorous agitation potassium permanganate (1.5 g) was slowly added to the suspension by carefully controlling temperature less than 20 °C. Then, the ice bath was removed and temperature was allowed to rise to 35 ± 5 °C and maintained for half an hour and the mixture appears as a thick paste. At the end of further 20 minutes, 25 mL water was added and left for 15 minutes. The temperature rises to 90 \pm 5 °C during addition of water. Then, 100 mL water was added for further diluting the solution. 30% H₂O₂ solution was subsequently added to it and the color became bright yellow. The solution was then filtered in warm condition to avoid precipitation. The residue was collected and further washed several times. The graphite oxide so obtained was dispersed in water and sonicated with about 25 kHz frequency and power of 650 W for 1 hour resulting information of a clear brown dispersion of graphene oxide.

XRD analysis

X-ray diffraction measurements were performed using Bruker D8 Advance X-ray diffractometer operated at a current of 40 milli Amp and 40 kV voltage. The scan rate was 2⁰/minute in the 20 range of 200–900 during data accumulation. The Cu K α (λ = 0.1546 nm) was used as the radiation source for the experimental measurements.

Transmission Electron Microscopy (TEM) measurement

TEM images were taken on electron microscopes (JEOL-2010 and FEI Tecnai S-twin) operated at an acceleration voltage of 200 kV. A drop of aqueous solution of GO and SG were placed on a carbon-coated copper grid of 400 mesh and allowed to soak. Then, the grids were kept overnight in vacuum desiccator to dry properly.



Fig. S1 XRD spectrum of GO.



Fig.S2 TEM image of GO.



Figure S3 The FAD fluorescence emission spectra of blank GOX (166 μ g/mL) (a) and in presence of GO of concentrations 3.32 (b), 6.62 (c), 9.9 (d), 13.16 (e), 16.4 (f), 19.6 (g) and 26 (h) μ g/mL in 0.1 M PBS (pH 7.0) under excitation wavelength 450 nm. Inset shows Stern-Volmer plot of fluorescence quenching of FAD of GOX by the addition of GO.



Figure S4 Circular Dichroism spectra of blank GOX (0.5 mg/mL) and in the presence of various concentrations of GO in near- UV region.



Fig. S5 Michaelis-Menten plot of blank LDH and in presence of different inhibitor (GO) concentrations of 0 (a), 0.02 (b) and 0.2 μ g/mL (c).



Fig. S6 Lineweaver-Burk plot of the GOX blank and in presence of GO of concentrations 0 (a), 0.02 (b) and 0.2 μ g/mL (c).

Reference

1. W. S.Hummers and R. E. Offeman, J. Am. Chem. Soc., 1958, 80, 1339–1339.