Electronic Supplementary Material (ESI) for RSC Advances.

ROS generation by reduced graphene oxide (rGO) induced by visible light showing antibacterial activity: comparison with graphene oxide (GO)

Supporting information

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1. Materials and Instrumentation

Graphite powder (50<μm, with purity of 99.5wt%, CDH Fine Chemicals), sodium nitrate (NaNO3), conc. H2SO4, potassium permanganate (KMnO4), hydrogen peroxide H2O2(30%v/v), HCl, were procured from Merck India Ltd. hydrazine hydrate 80%,sodium borohydride (NaBH4), hypo phosphorous acid(H3PO2), sodium dithionite(Na2S2O4) SPECTROCHEM India Ltd. Borosilicate Teflon lined screw capped culture tubes were procured from Borosil. Electronic absorption spectral measurements were performed in a Jasco V-630 UV/Vis Spectrometer. Systronics^{R} Digital pH meter 335 was used to measure the pH of the solution. Lab Man Scientific Instrument ultrasonic cleaner LMUC-4 was used for sonication while RM-12C micro centrifuge was used for centrifugation. The IR spectra were recorded using a JASCO FT-IR –460 plus infrared spectrometer in the 4000 – 400 cm^{-1} frequency range, using powered samples diluted in KBr pellets. For fluorescence image Nikon inverted microscope (Nikon eclipse Tí, Japan) at 60x objective lens was used.

2. Method

GO has been prepared using Hummers method [1].

3. Reduction of GO:

Four different reducing agents have been used to reduce the GO prepared by modified Hummers method.

a) N2H4 method

Reduction was done as reported earlier [2].

b)H3PO2 method

It was reduced as per the reported method

c) NaBH4 method

NaBH4 was used to reduced GO according to the reported method [4].

d) Na2S2O4 method

Earlier reported method was utilized to reduce GO [5].

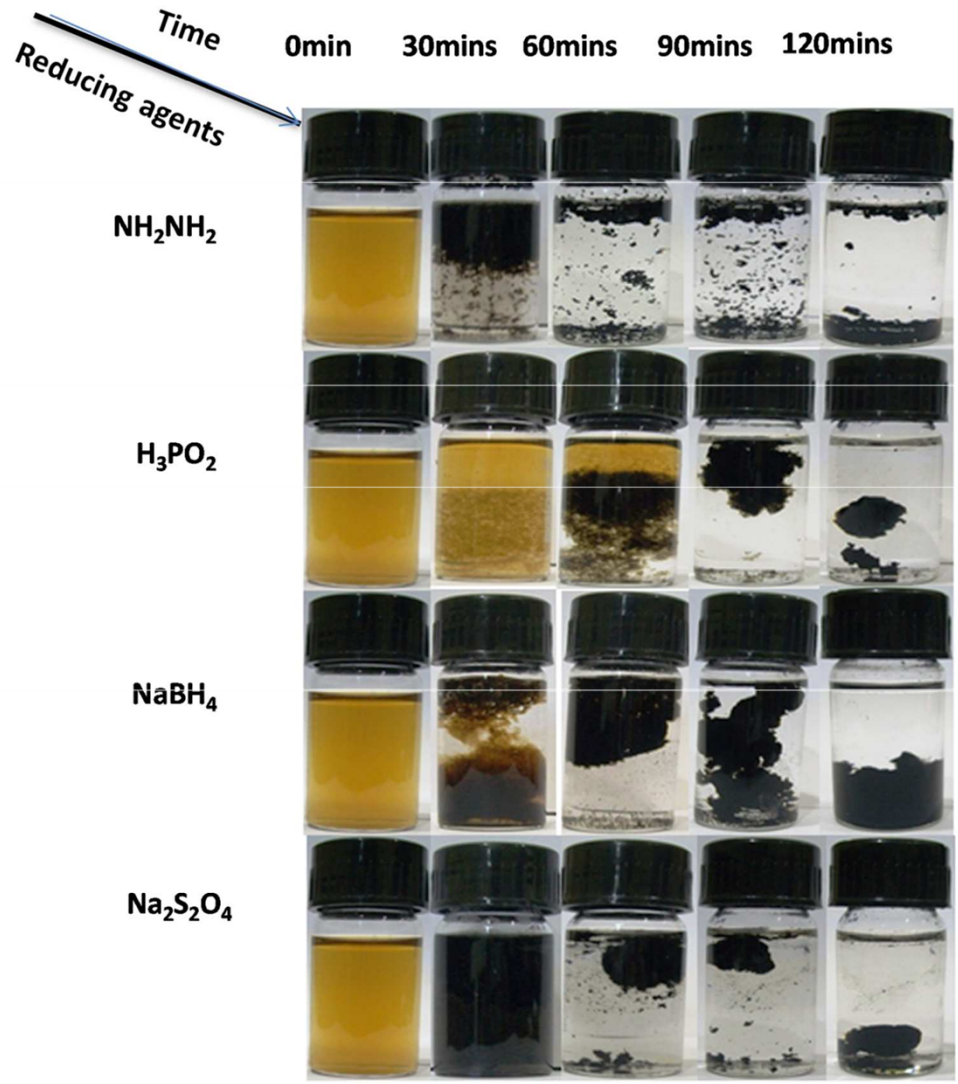

Figure S1 Shows the gradual reduction of GO to rGO with various reducing agents with time at an interval of 30 mins.

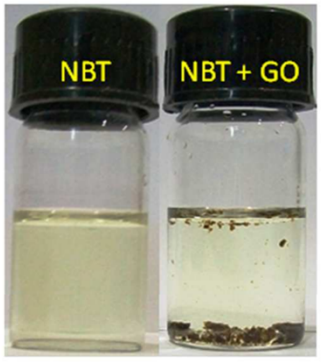

Figure S2 Shows **no** blue colour formation using GO under visible light exposure , in 10mL of 0.6µmol NBT solution GO (5mg) .

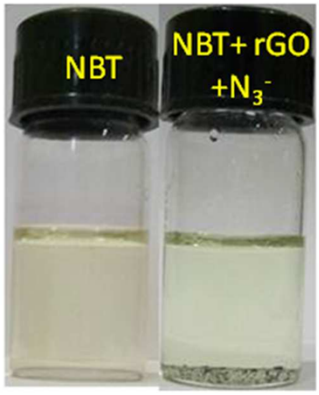

Figure S3 Shows **no** blue colour formation in 10mL of 0.6µmol NBT solution rGO (2 mg) and sodium azide(1mg) under visible sunlight.

4. pH metric experiment:

2 mg. rGO was dispersed in 10 mL of 0.6µmol NBT solution by stirring for 5 minutes and the pH was monitored. The mixture was exposed to sunlight and after 15 minutes of interval the pH of the light exposed solution was measured. In the presence of light the pH of solution steadily increased but after 90 minutes the rate of increment slowed down considerably and finally remained unchanged.

5. Catalytic experiment:

2 mg. rGO dispersed in 10 mL of 0.6µmol NBT solution was stirred for 5 minutes and exposed under sun light. After one hour the yellow colour of the solution changed to blue due to the formation of diformazan. The mixture was centrifuged and the supernatant is subjected to electronic spectral measurement fixing the optical density at 588 nm to measure the formation of blue diformazan . The residue was washed thoroughly with de- ionized water and the light induced experiment with this used rGO in the presence of fresh NBT was repeated. After several such cycles the activity of the rGO is diminished. On washing the finally treated rGO with dilute alkali the GO formed on the surface gets

solubilized and the alkali leached solution shows fluorescence but the residue remains non- fluorescence retaining the unused rGO.

6. References:

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4. J. Li, H. Lin, Z. Yang and J. Li, *Carbon*, 2011, **49**, 3024-3030.
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