Electronic Supplementary Material (ESI) for Journal of Materials Chemistry B.

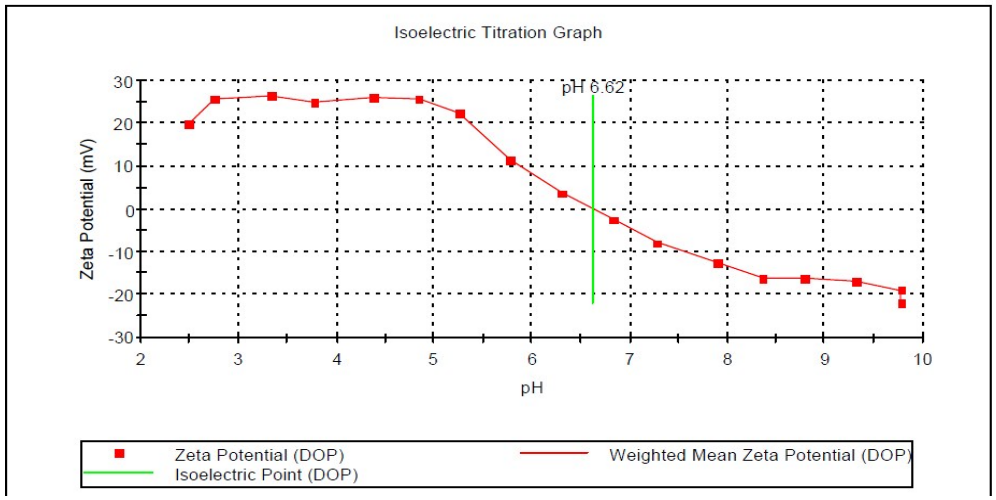
Supporting Information

Blood Brain Barrier Permeable Gold Nanocluster for Targeted Brain Imaging and Therapy: An *in vitro* and *in vivo* Study

*L.V.Nair, R.V.Nair, S.J.Shenoy[#], A.Thekkuveetil[+], R.S.Jayasree**

[*]Dr. R. S. Jayasree
Division of Biophotonics and Imaging ,[#]Division of in vivo models and Testing,[+] Division of Molecular Medicine
Sree Chitra Tirunal Institute for Medical Sciences andTechnology (SCTIMST), Trivandrum-695012
Tel: +91-471-2520273
Fax: (+)91-471-2341814
E-mail: jayasree@sctimst.ac.in, jayashreemenon@gmail.com

a)



b)

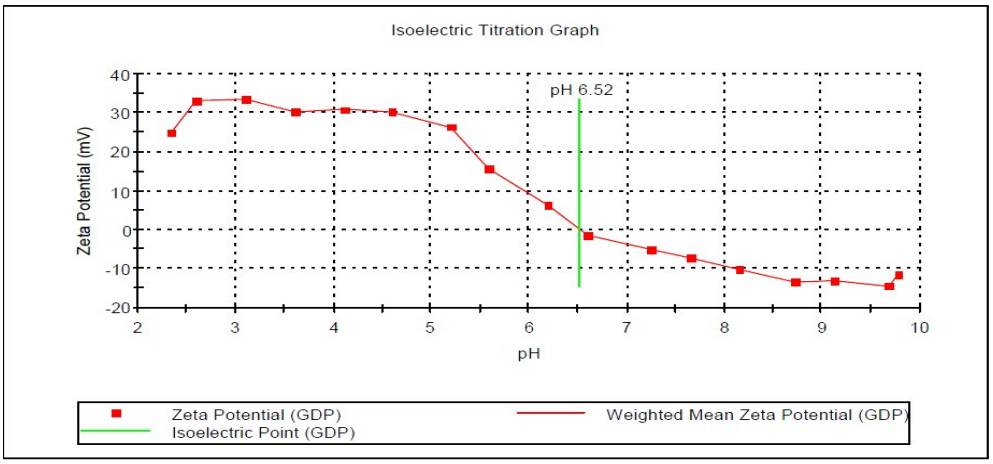


Fig S1: Isoelectric point measurement using zeta sizer a) L- Dopa and b) Dop@GQC.

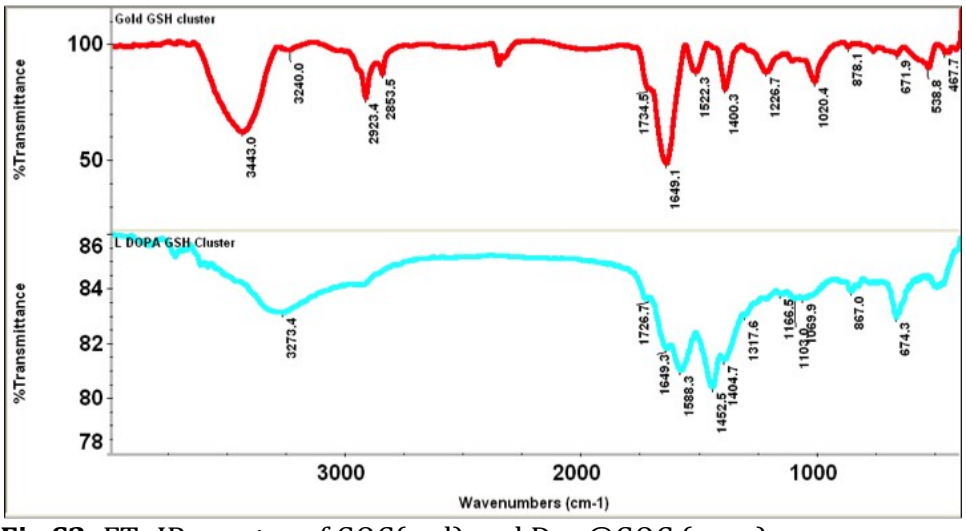


Fig S2: FT- IR spectra of GQC(red) and Dop@GQC (cyan)

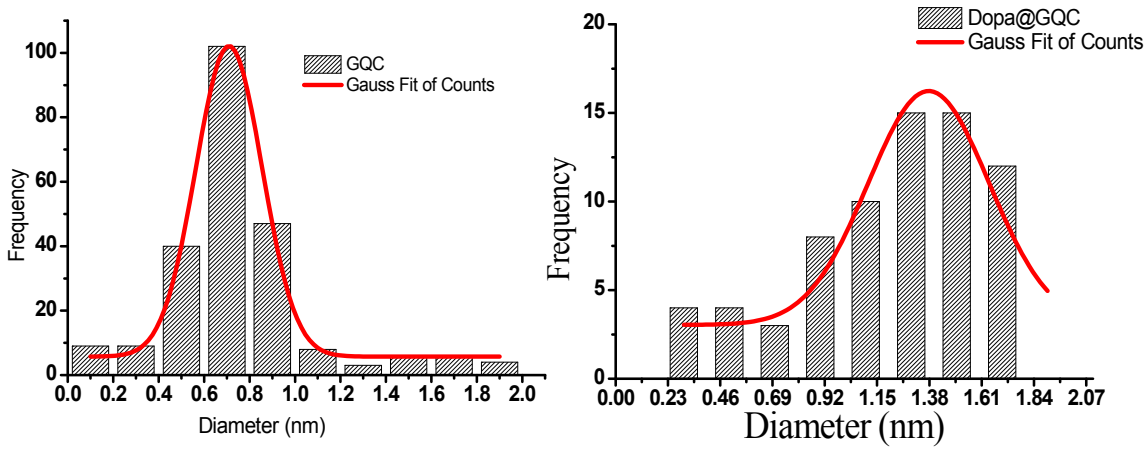


Fig S3: Particle size distribution of GQC and Dopa@GQC

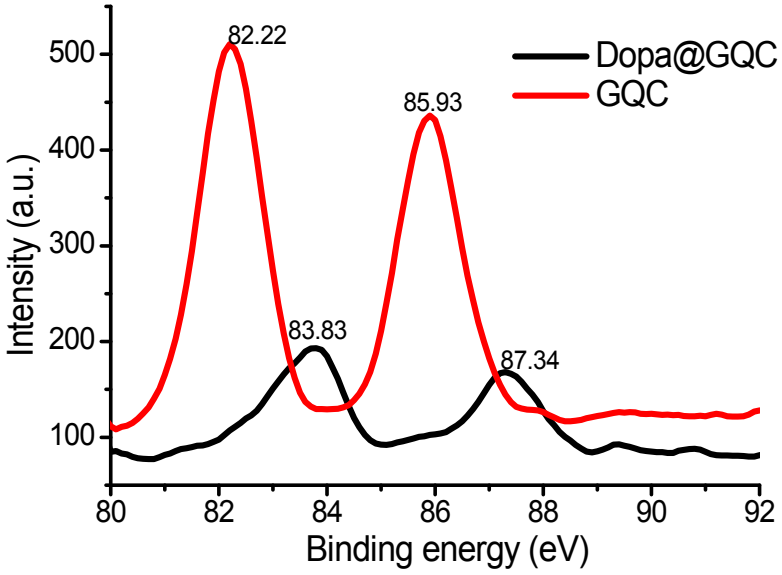


Fig S4: XPS spectra of GQC and Dop@GQC

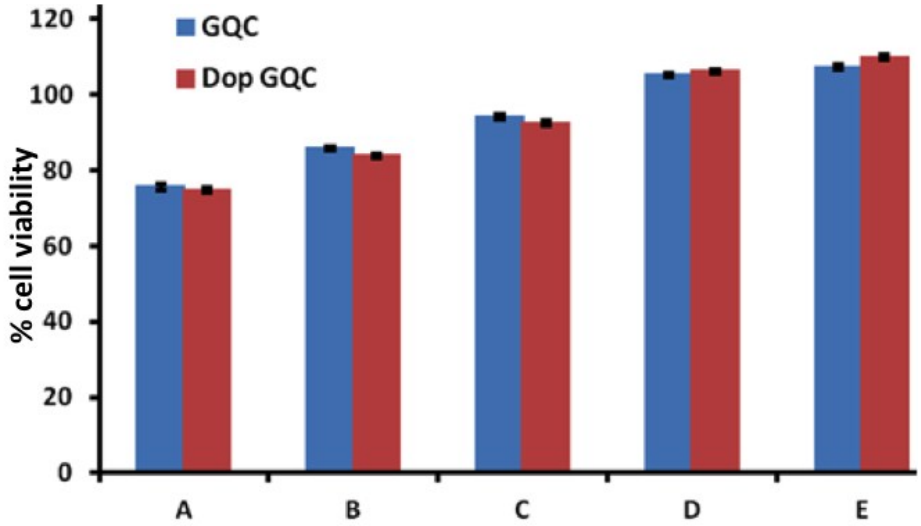


Fig S5: Viability of the cells by the addition of different concentration of materials using MTT assay. A, B, C, D, and E represents 1, 0.5, 0.1, 0.05 and 0.01 mg/ mL of material respectively.

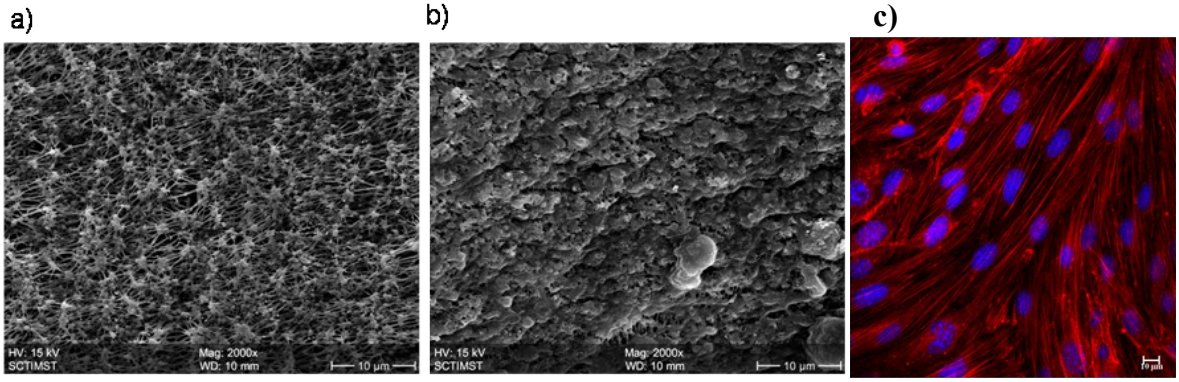


Fig S6: ESEM images of a) Milli cell Insert b) bEnd.3 cell monolayer on forming tight junction c) Actin staining of bEnd.3 monolayer tight junction.

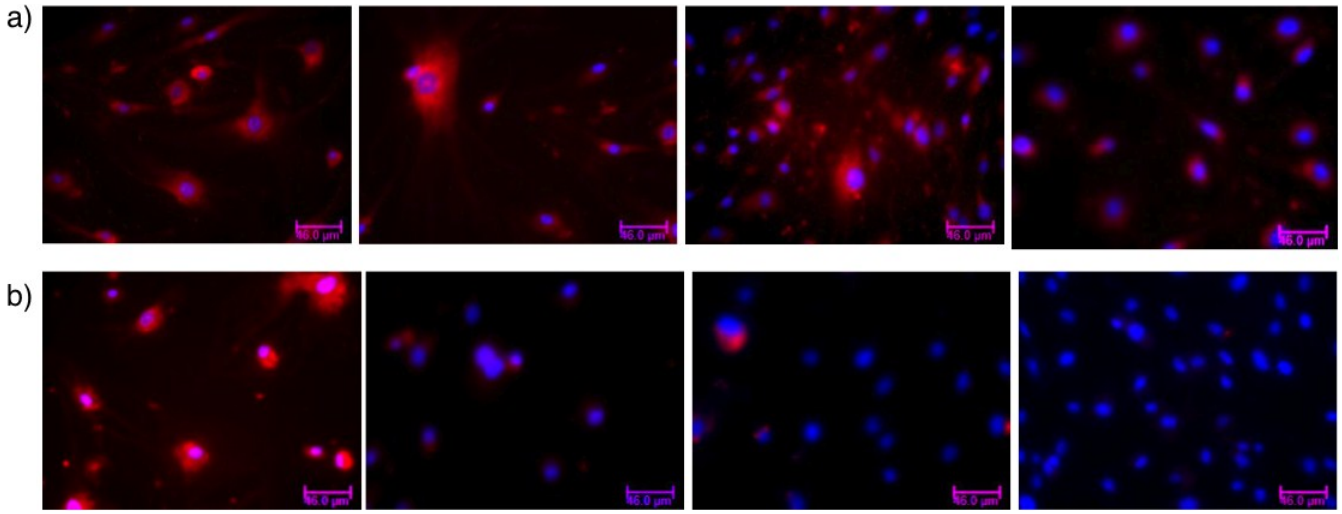


Fig S7: Cellular uptake of the material for 3, 6, 12 and 24 h using fluorescence microscope. a) Bend3 cells with GQC for 3 h, 6 h, 12 h and 24h b) represents the same with Dopa@GQC.

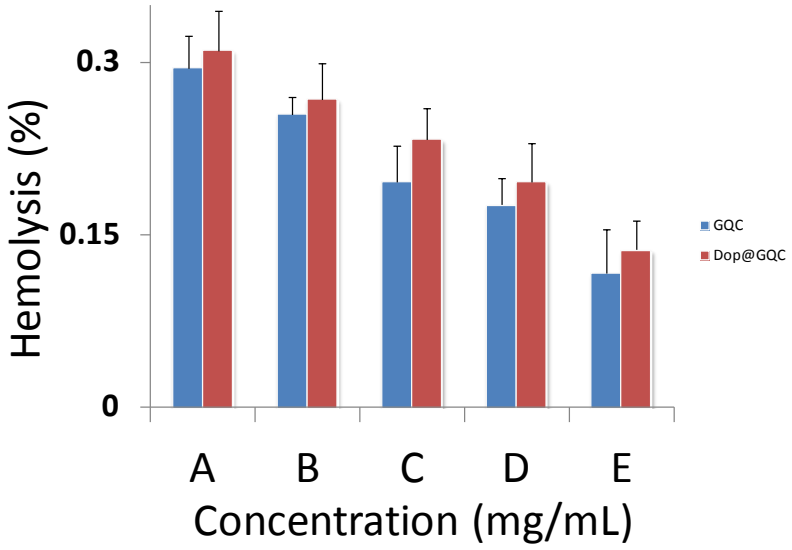


Fig S8: Percentage hemolysis of GQC and Dopa@GQC. A, B, C, D, and E represents 1 mg, 0.5 mg, o.1 mg, 0.05 mg, and 0.01 mg/ mL of material respectively.

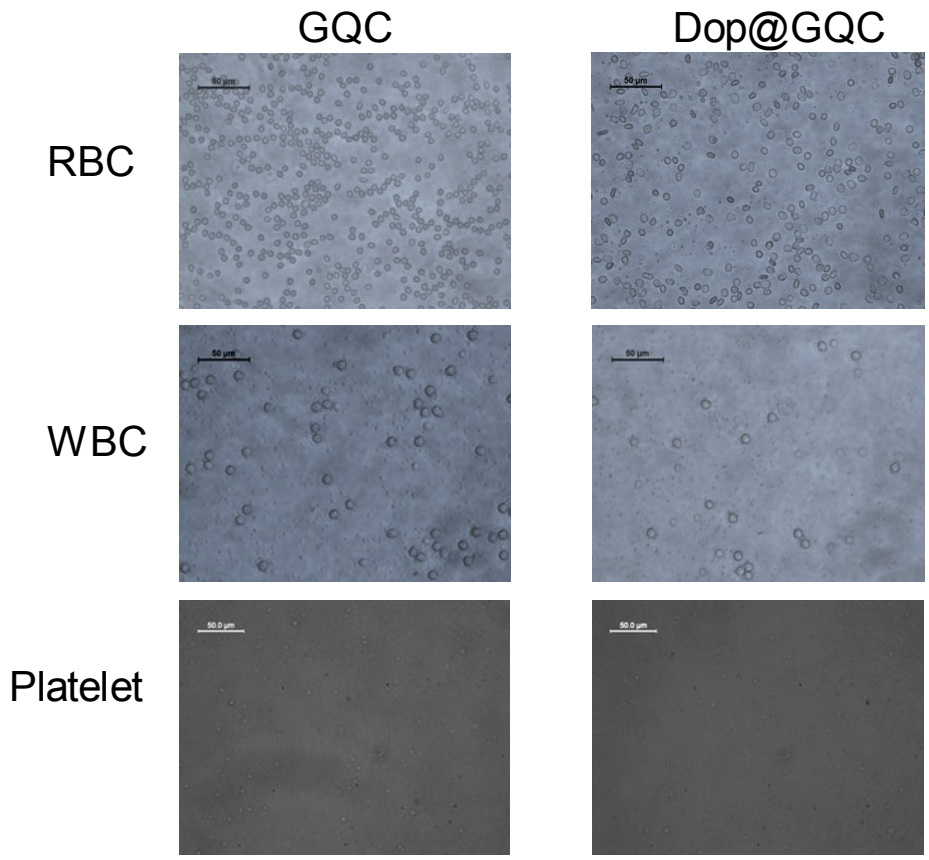


Fig S9: Response of blood cells towards the nanoparticles.

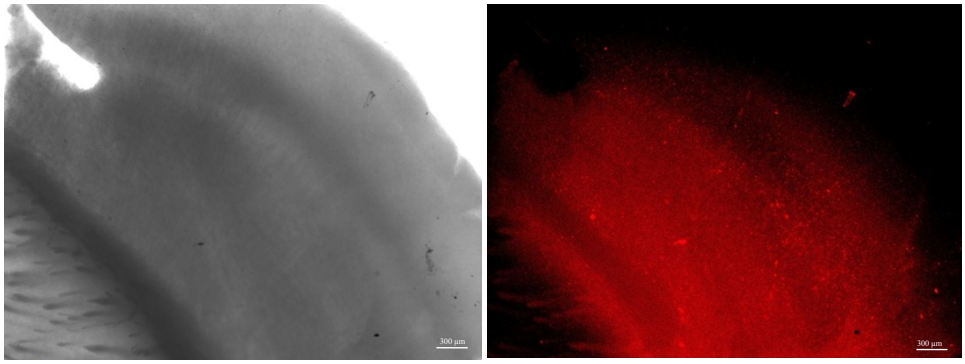


Fig S10: Bright field and fluorescence microscopic images of brain section of Dopa@GQC injected animal showing fluorescence emission of the gold cluster from the brain (Both white and grey matter are visible).

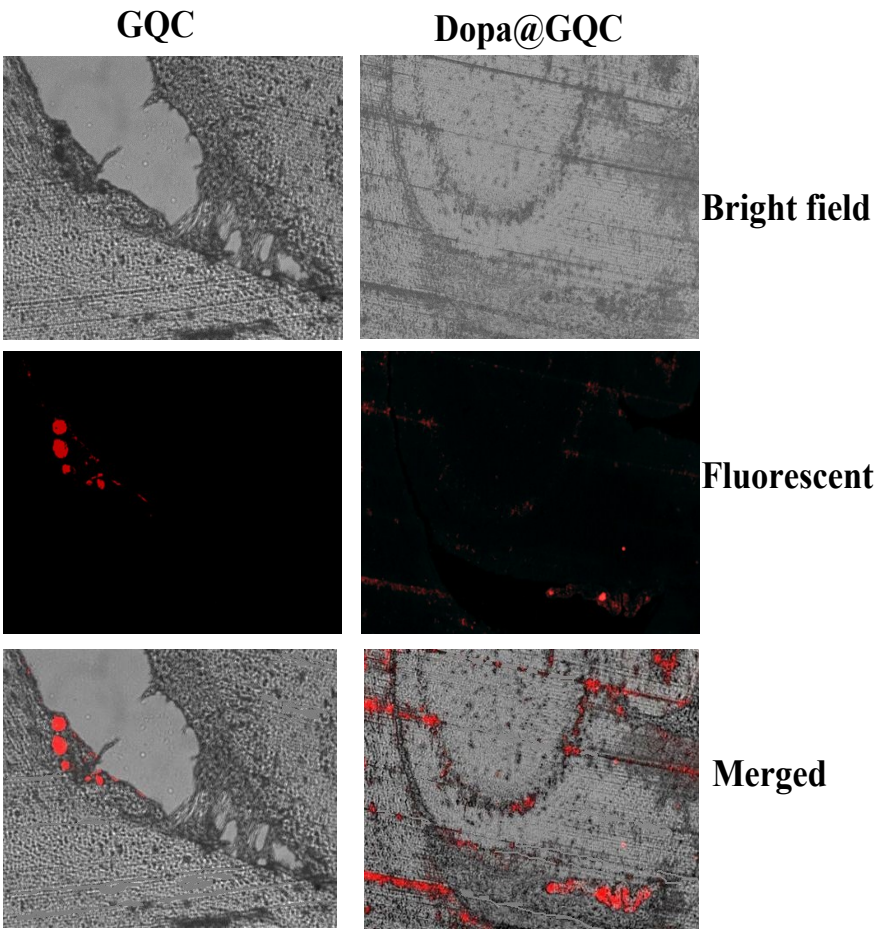


Fig S11: Images of brain sections showing the fluorescence in the case of Dopa@GQC and GQC. The confinement of GQC inside the blood vessel is clear in the case of GQC. In the case of Dopa@GQC, bright fluorescence from areas of hippocampus is very clear.

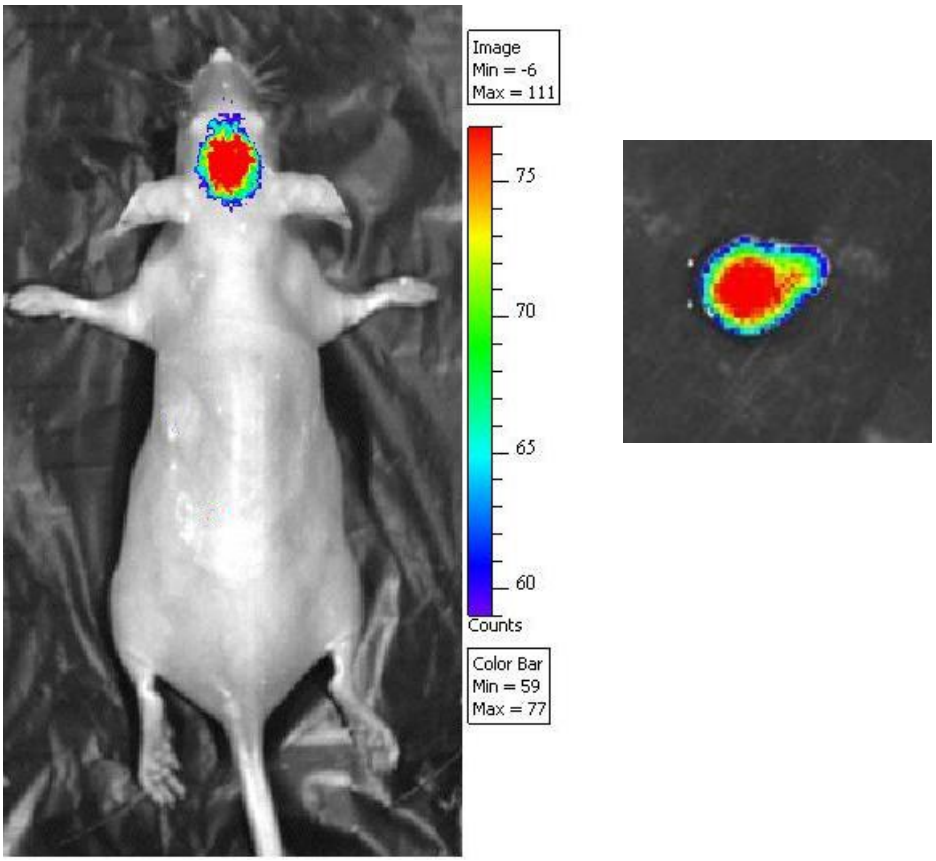


Fig S12 In vivo and ex vivo images of animal injected with dopa@GQC after saline perfusion

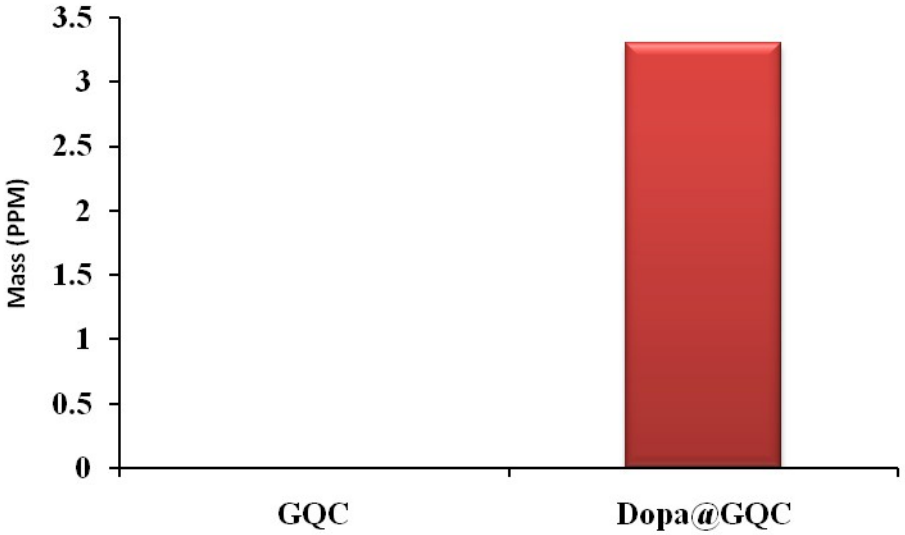


Fig S13: ICP-OES analysis results of of GQC and Dopa@GQC

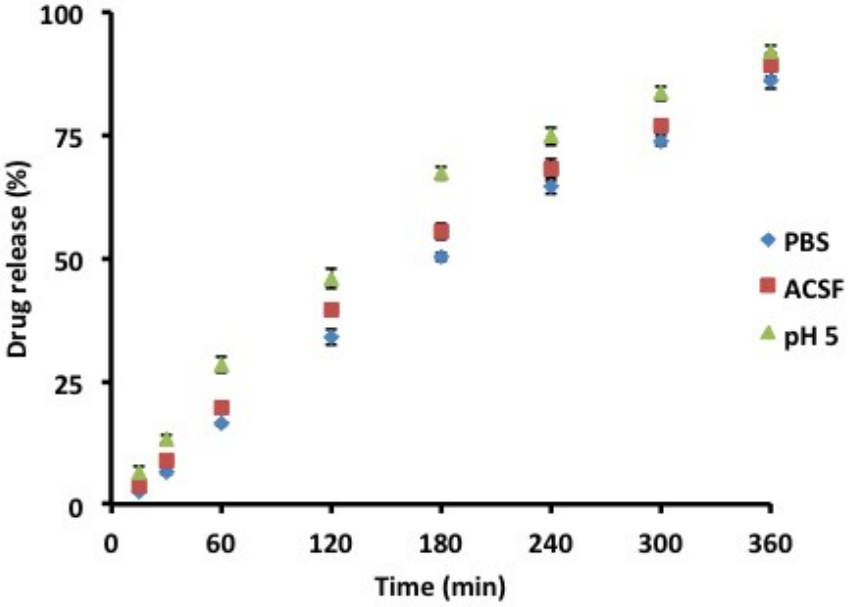


Fig S14: Cumulative pilocarpine release profile of pilocarpine-Dopa@GQC in PBS (pH 7.3), ACSF and phosphate buffer (pH5).

Scanning Electron Microscopy (SEM) of b End.3 cell

To confirm the complete monolayer formation of bEnd3 cells, SEM study was carried out after 6th day of cell seeding. Briefly, after 6th day the media in the culture insert were removed and washed twice with PBS. 1% formaldehyde was added to the insert to fix the cells and kept in 4^{o}C for 12 h. Cells were dehydrated with different grades of alcohol. After critical point drying and gold coated cells were viewed under SEM (Hitachi, 2400). Millicell insert without cells and with media were served as a control.

Visualization of tight junctions-Actin staining

The cells were cultered and then permeabilised using 0.2% Triton X-100 in blocking solution, made of 1% (w/v) bovine serum albumin (BSA) in PBS, for 20 min.
The permeabilised cells were then washed twice with PBS and incubated with 250 µl of 1% BSA for 30 min.
The blocking solution is removed and cells were incubated with 200 µl rhodamine phalloidin solution (0.2 µg/ml) for 20 min at room temperature. After removal of rhodamine phalloidin, the cells were treated with 1% BSA as before.
The cells were washed with PBS, and dried overnight at 4°C. Images were obtained using inverted confocal laser scanning microscope, equipped with He/Ne laser 543. The visualization of rhodamine phalloidin was done using excitation and emission wavelengths of 543 and 605 nm, respectively.