

Supporting Information

Emergence of fluorescence in boron nitride nanoflakes and its application in bioimaging

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FESEM images

FESEM images of BNNFs, sample was prepared by putting the drop on the glass slide of dispersed sample by ultrasonication for 30 min. It is clear from the images that 30 min ultrasonication also could not get rid of agglomeration due to high surface energy between nanoflakes.

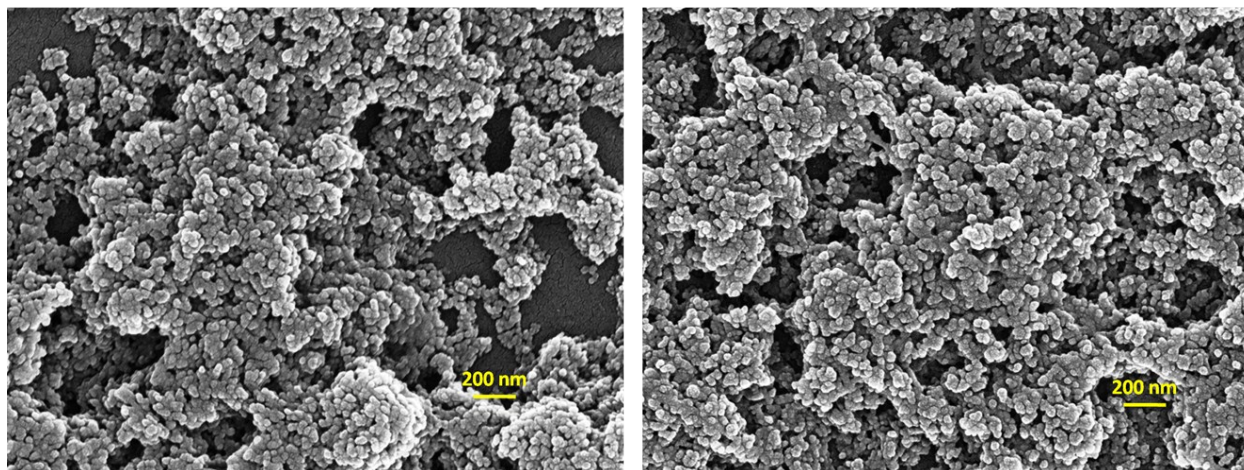


Figure S1. FESEM images of the sample showing the high agglomeration between the flakes due to high surface energy.

X-Ray Diffraction

X-Ray diffraction for as prepared material was also performed in range 10° - 90° with a rate of $1^{\circ}/\text{min}$ and found to closely match with hexagonal boron nitride (h-BN), JCPDS reference 85-1068. Indexing of the pattern was shown in the figure 2.

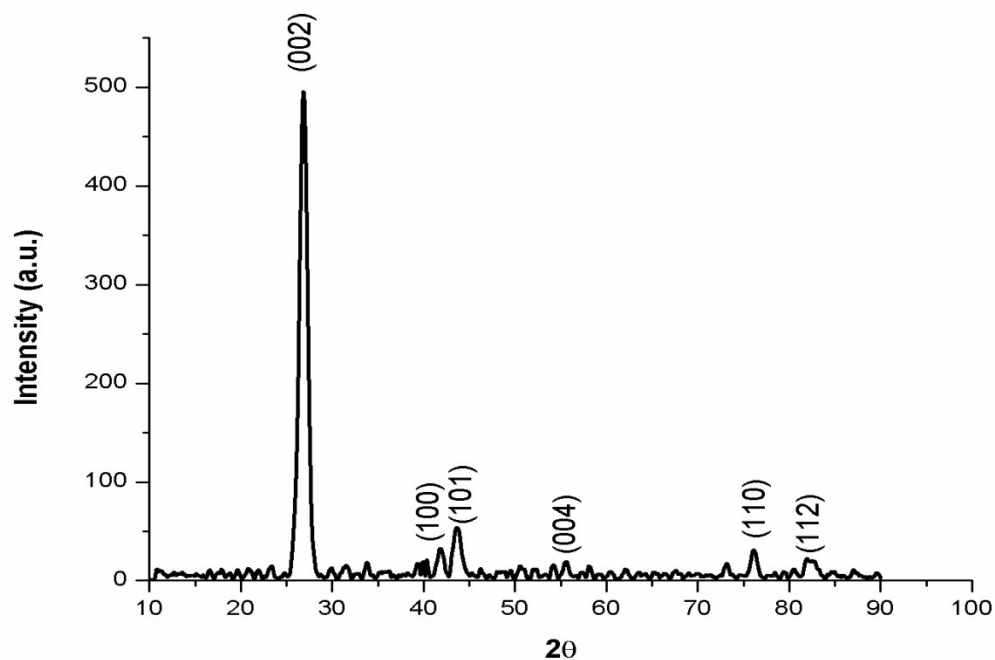


Figure S2: XRD pattern of as prepared materials closely matches with hexagonal boron nitride (h-BN).

Size distribution by Zeta Sizer

Size distribution study was performed by Zetasizer Nano ZS90, showing that a good fraction of the size of the nanoflakes was distributed over 20-40 nm by volume.

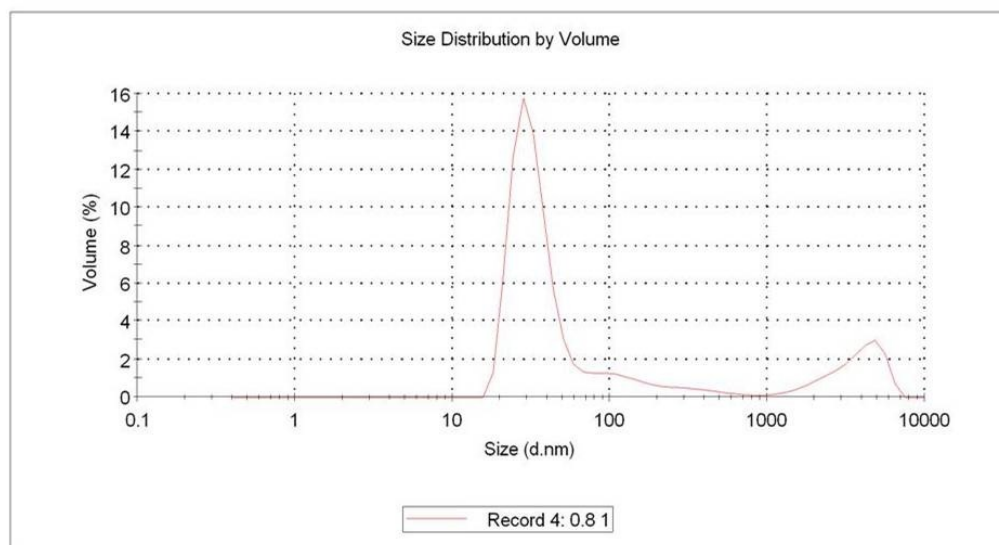


Figure S3. Zeta size distribution by volume

Fluorescence Spectra under the excitation at 243 nm and upon annealing in air

Fluorescence spectra at excitation 243 nm was recorded and emission was found that no significant change in the fluorescence spectra figure 3. It can be concluded that the fluorescence emission due to 243 nm excitation was overlapping with the existing emission due to BO_2^- species and zigzag structure at edges. Fluorescence of sample after annealing at 1000 °C in presence of air was recorded.

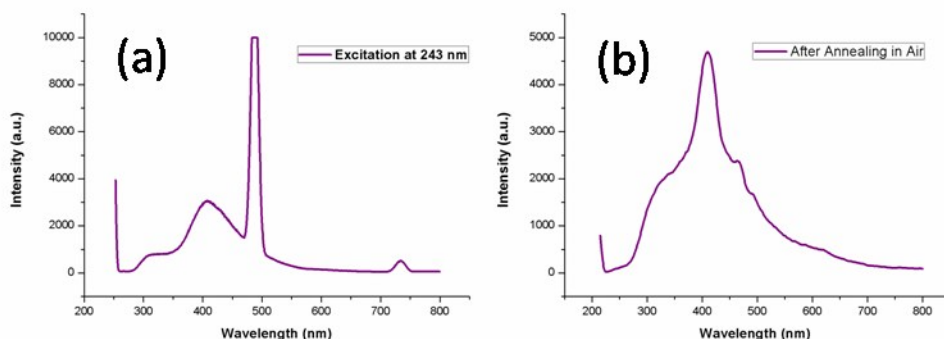


Figure S4. (a) Fluorescence excitation spectra with excitation at 243 nm; the main peak (at 415 nm) was having no shift. The peak at 486 nm is actually due to doublet excitation which is considered to be having no significance. (b) Fluorescence emission spectra for annealed sample in air at 1000 °C for 2 h.

Emission spectra was also recorded at an excitation wavelength 350 nm and it is observed that main peak at 415 nm remains unchanged and does not show any shift, while other peak at 435 nm become prominent. Peak at 435 nm has emerged as a sharp and dominant peak, which was observed as a hump in figure 4 (main article) as a result of BO_2^- species (as confirmed by FTIR spectra). Higher excitation wavelength will be favorable for more emission from BO_2^- species as compared to emission due to zigzag carbene structure. Emission spectrum at 350 nm excitation is given below.

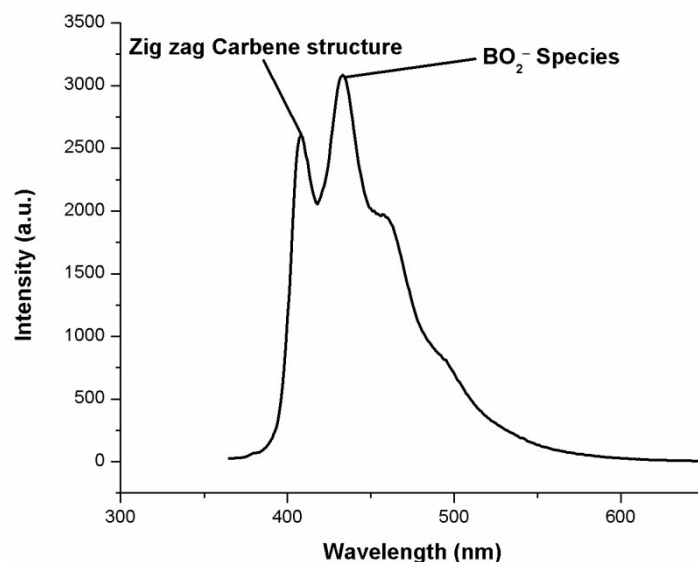


Figure S5: Excitation spectra at 350 nm excitation

Quantum Yield Measurements

To measure the quantum yield of the sample a single point method was performed using quinine sulphate as reference which is known to be having quantum yield of 0.57 at 350 nm as per previously reported method [1]. The mathematical equation to calculate quantum yield is given below

$$Q_S = Q_R \frac{I_S A_R \eta_S^2}{I_R A_S \eta_R^2}$$

Where Q represents the quantum yields, I refers to the integrated emission intensity (area under the curve); refractive index referred as η and A denote as the optical density. Subscript S and R stand for sample and reference respectively. Absorption of intensity was kept below 0.1, for measurement of emission using 10 mm fluorescence cuvette in order to minimize the re-absorption effect. The width of excitation slit and emission slit were kept 2.5 nm each for both to excite the samples and its fluorescence spectra was recorded in the wavelength range of 365-700 nm. The table 1 shows the calculated parameters from spectra.

Sample	Integrated emission intensity(I)	Absorbance at 350 nm (A)	Refractive index of solvent (η)	Quantum yield (Q)
Quinine sulphate	175832	0.0854	1.33	0.546
BNNFs	8936	0.096	1.33	0.025

Confocal microscopy images

Confocal microscopy images show that fluorescent BNNFs are internalizing into the cell as well as adhere on the cell surface. It can be attributed to higher degree of agglomeration as well as high interaction energy causing adhesion to cells surface (Figure S6 and S7).

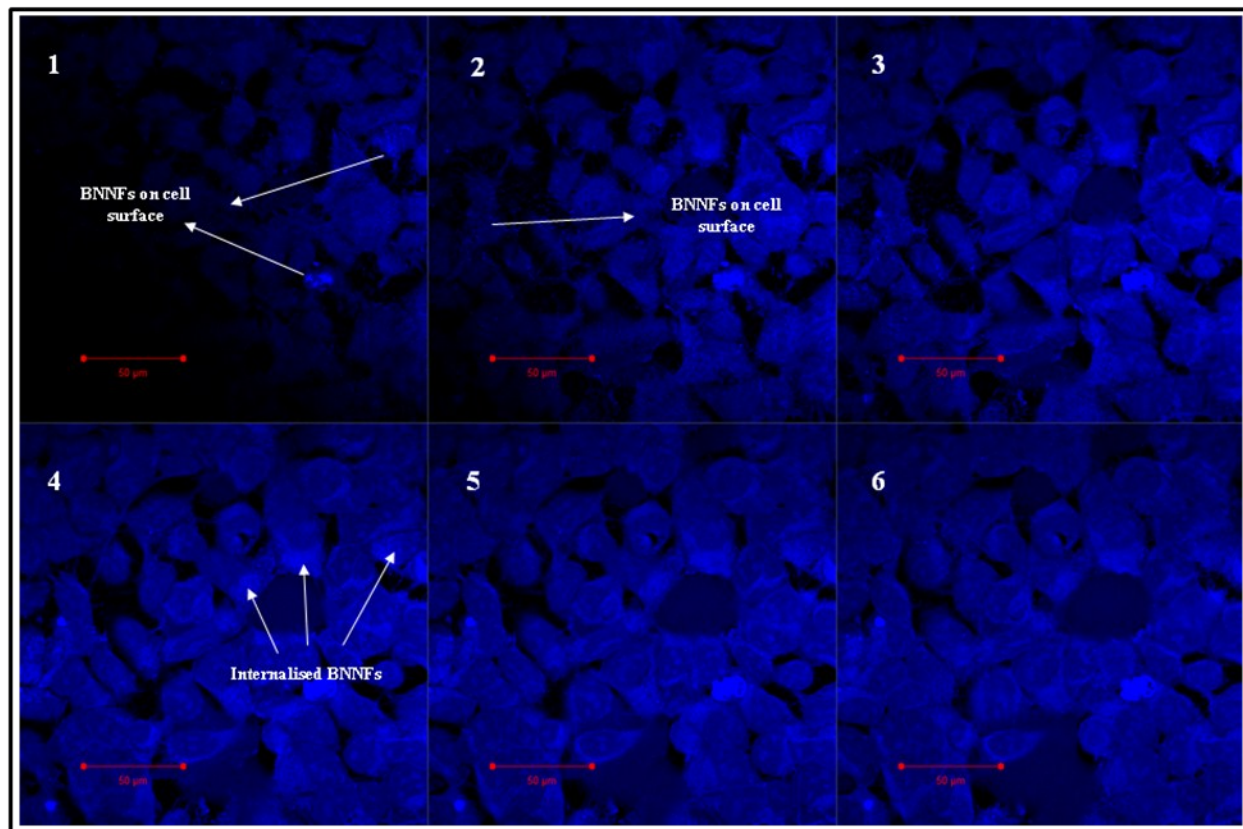


Figure S6: Z stack images of cells treated with BNNFs

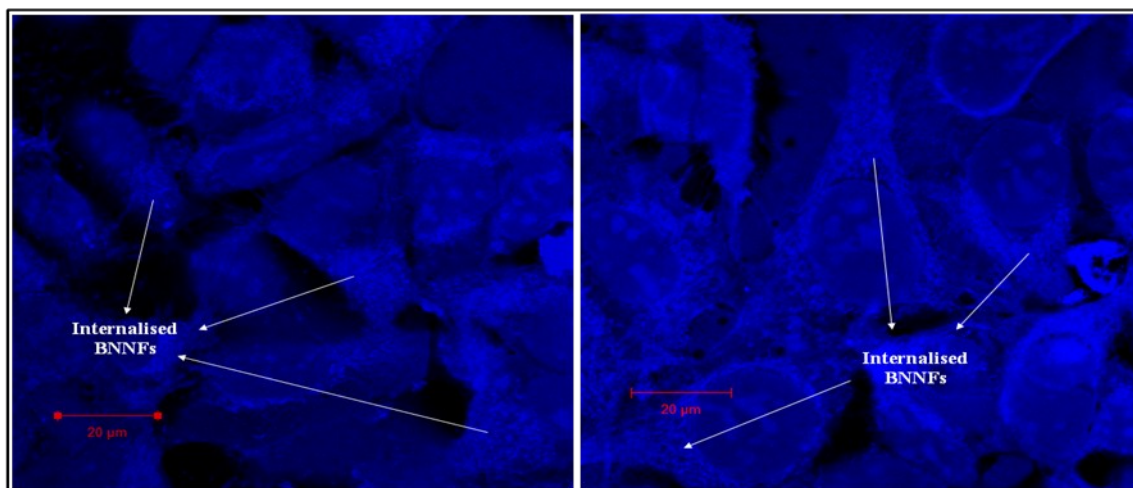


Figure S7: Internalized BNNFs inside MCF-7 cells higher magnification