ABSTRACT

Index No.: **160/18/Chem./26**

Thesis Title : **DESIGN AND DEVELOPMENT OF CARBON ALLOTROPE BASED SOFT MATERIALS FOR BIOSENSING AND BIOIMAGING**

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Nanobiotechnology is an extensively studied branch where amalgamation of biology and nanotechnology took place with purpose and objectives. Biomolecules in cellular environment execute their functions in marvelous precision and complexity. To understand this nano-dimensional network of functions, nanomaterials mimicking the function and properties of biomolecules open a new gateway of research for scientist. Among many kinds of nanomaterials, Carbon dot, also known as the carbon nanodot, carbon quantum dot, represents an interesting class of carbonaceous nanomaterial.The 'zero-dimensional' CDs have intrinsically fluorescent property, low toxicity, cytocompatibility and many other interesting physiohemical features.These remarkable characteristics mixed with their tunable surface emission property make them a potent candidate for chemical sensing, bioimaging, drug carrier and also a theranostic agent themselves. To this end, in this thesis I report, judicious designing of carbon dot for photodynamic therapy, combination therapy and hypoxia assisted apoptosis against cancer cells along with the development of carbon dot based guanine sensor.

Chapter 1 describes a photosensitizer (riboflavin) tailored surface functionalized carbon dot (**RCD1s**) for visible light induced targeted cancer therapy. Phenylboronic acid appended biotinylated blue emitting carbon dot (**CD1s**) was linked with Riboflavin to prepare green emitting **RCD1s** by using complementary boronate-diol linkage. This newly synthesized **RCD1s** has the ability to produce reactive oxygen species (ROS) such as hydroxyl and superoxide radicals under exposure of visible light (wavelength:460−490 nm). Thus, under irradiation of visible light (wavelength: 460−490 nm), **RCD1s** was found to kill HeLa and B16F10 melanoma cells over non-cancer cellNIH3T3 by ~5-fold higher efficacy through ROS induced oxidative DNA damage. The presence of biotin on the surface of the riboflavin tethered carbon dot is essential for the selective killing of cancer cells over normal cells. Besides, **RCD1s** in the presence of visible light selectively stained HeLa and B16F10 cells over noncancerous cell NIH3T3 by exploiting its fluorescence and cancer cell targeting moiety, biotin. Hence, the newly developed **RCD1s** can be utilized in theranostic applications including bioimaging and selective killing of cancer cells in the presence of visible light (460−490 nm) (Figure 1).

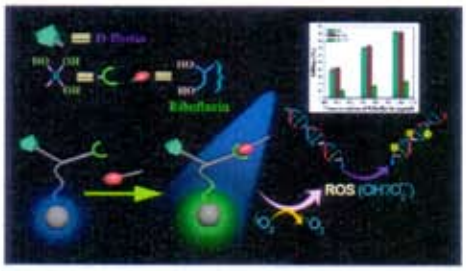

Figure 1

Chapter 2 delineates the design and preparation of covalently tailored biotinylated Fe^{2+}-doped carbon dots (**FCD_b**). The **FCD_b** was successfully used as a pro-drug activator, diagnostic probe, and target-specific delivery vehicle for anticancer drug paclitaxel in pro-drug-free drug combination therapy of cancer treatment. Fe^{2+}-doped carbon dot (**FCD**) was covalently modified with cancer cell targeting ligand biotin (**FCD_b**). **FCD_b** emit blue fluorescence under UV light irradiation. **FCD_b** can effectively sense H_2O_2 by fluorescence quenching as well as activate H_2O_2 (pro-drug), which oxidatively damage the DNA through the generation of reactive oxygen species (ROS: superoxide, hydroxyl radical etc. **FCD_b** was utilized as selective cellular markers for cancer cell B16F10 owing to their high H_2O_2content, which was more distinct due to the overexpression of biotin receptor in cancer cell. Anticancer drug paclitaxel (PTX)-loaded **FCD_b** (**FCD_b-PTX**) was exhibited ~2.7- to 3.5-fold higher killing of B16F10 cells in comparison to non-cancer NIH3T3cells through the synergistic action of ROS and anticancer

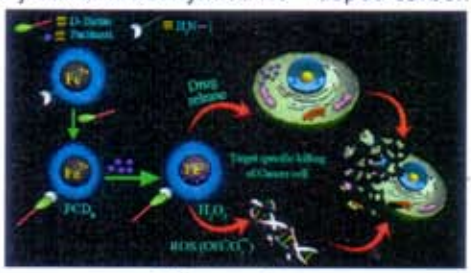

Figure 2

effect of PTX. Hence, this newly developed pro-drug-free drug agent **FCD_b-PTX** can act as a potential theranostic agent in the domain of combination therapy of cancer treatment (Figure 2).

Chapter 3 deals with biotinylated Co^{2+}-integrated blue emitting carbon dot (**$CoCD_b$**).Conventional cancer treatments have systematic side effects that stand against its desirable therapeutic efficacy. Alternative strategies using biochemical features of cancer cells to promote apoptosis are finding notable significance. One such important biochemical feature of malignant cells is hypoxia, alteration of which can lead to cell death. Hypoxia inducible factor 1α (HIF-1α) has the key role in hypoxia generation. Herein, we synthesized **$CoCD_b$** that specifically diagnose and selectively killed cancer cells with ~3-3.1 fold higher efficiency over non-cancer cells by hypoxia induced apoptosis in absence of traditional therapeutic intervention. Immunoblotting assay in **$CoCD_b$** treated MDA-MB-231 cells confirmed the increased expression of HIF-1α that was responsible for efficient killing of cancer cells. In 2D cells and 3D tumor spheroid, **$CoCD_b$** treated cancer cells showed significant apoptosis that make **$CoCD_b$** a potential theranostic agent (Figure 3).

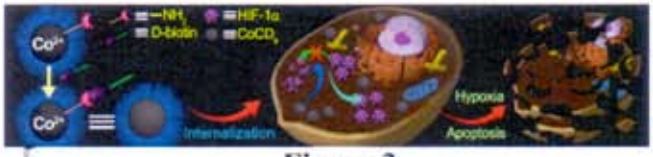

Figure 3

Chapter 4 demonstrates development of copper (Cu^{2+}) doped carbon dot (**CuCD**) via hydrothermal method. This carbon dot exhibited blue emitting excitation dependent emission at 428 nm when excited at 340 nm. TEM, AFM images confirmed the size of **CuCD** ~5 nm. Deconvulated XPS spectra of **CuCD** revealed the presence of Cu^{2+} in the synthesized carbon dot. We explored fluorescent turn-off method using the prepared **CuCD** as probes to realize a simple, sensitive and selective analysis of guanine. The fluorescence intensity of **CuCD** got quenched significantly in presence of guanine due to the formation of a stable interaction between Cu^{2+} ions and guanine. Accordingly, the **CuCD** is used as fluorescence probes for application in the detecting guanine having the limit of detection (LOD) = 1.57 μM. Moreover, along with the adenine, thymine, cytosine, different biomolecules and metal ions that are abundant in cellular environment have no interference for the detection of guanine by **CuCD**. In cellular studies, this **CuCD** can efficiently quenched guanine treated cell line (B16F10 and NIH3T3) with respect to the control untreated ones and having cell viability upto ~90%. So, therefore a highly sensitive and selective ratiometric fluorescence probe **CuCD** for guanine detection was synthesized which was also utilized for bioimaging (Figure 4).

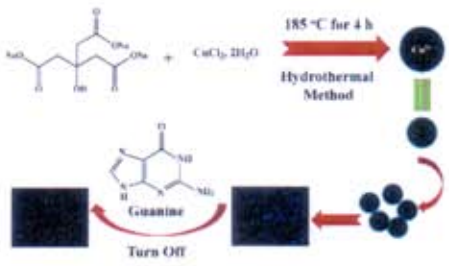

Figure 4

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