Nanobio Hybrids for Stimuli Transduction

Elena A. Rozhkova[1] , Ilya Ulasov[2], Maciej S. Lesniak[2] Nada M. Dimitrijevic [1], Barry Lai[3], Valker Rose[3], and Tijana Rajh[1]

[1] *Center for Nanoscale Materials, Argonne National Laboratory*
[2] *University of Chicago*
[3] *Advanced Photon Source, Argonne National Laboratory*

Scientific Trust Area: Design of functional materials at nanoscale

Research Achievements: Functional nanoscale materials that possess specific physical or chemical properties are able to leverage signal transduction *in vivo*. Once these hard materials integrated with biomolecules they combine properties of both inorganic and bioorganic moieties for successful interfacing with whole cells for direct manipulation and changing biochemical pathways via energy or information transmittance. These systems are appealing for wide range of application from the life sciences to advanced catalysis and clean energy production.

Semiconductor metal oxide particles such as TiO_2 have been of interest in a wide range of applications including dye-sensitized solar cells, gas sensors, photocatalytical degradation of organic substrates and deactivation of microorganisms. TiO_2 is a semiconductor with a bulk band gap of 3.2 eV that can induce reactive oxygen species (ROS) in aqueous medium under UV light exposure. Our strategy in construction of TiO_2 based nano-bio hybrids is based on application of natural dihydroxybenzenes (e.g. dopamine, DOPAC, L-Dopa) as linkers, which due to presence of two OH- groups in the ortho- position form a strong bidentate complex with coordinatively unsaturated Ti atoms at the surface of nanoparticles. When biomolecules, such as DNA or proteins are covalently bound to dopamine, this linker acts as a conductive bridge between TiO_2 nanocrystals and biomolecules allowing transport of photogenerated holes to biomolecules. Modification of TiO_2 nanoparticles with dopamine enables harvesting of visible light, and promotes spatial separation of charges. The formation of oxidative species (OH, $^1O_2$, O_2^-, HO_2, H_2O_2) upon illumination of TiO_2-dopamine was studied using complementary spin-trap EPR and radical-induced fluorescence techniques. The localization of holes on dopamine suppresses oxidation of adsorbed water molecules at the surface of nanoparticles, and thus formation of OH radicals. At the same time, dopamine does not affect electronic properties of photogenerated electrons and their reaction with dissolved oxygen to produce superoxide anions [1]. In experiments *in vivo* we demonstrated that under visible light illumination TiO_2-DOPAC-antibody system catalyses generation of superoxide, a relatively long-lived ROS; this ROS serves as a secondary messenger altering of cellular respiratory pathways for re-programming cancer cells intrinsic death agenda, Figure 1.

Of equal significance, for first time we report direct visualization of ligand-receptor interaction and mapping of a specific human GBM receptor through a single brain cancer cell using TiO_2 nanoparticles through XFM hard X-rays of the Advanced Photon Source, Figure 2.

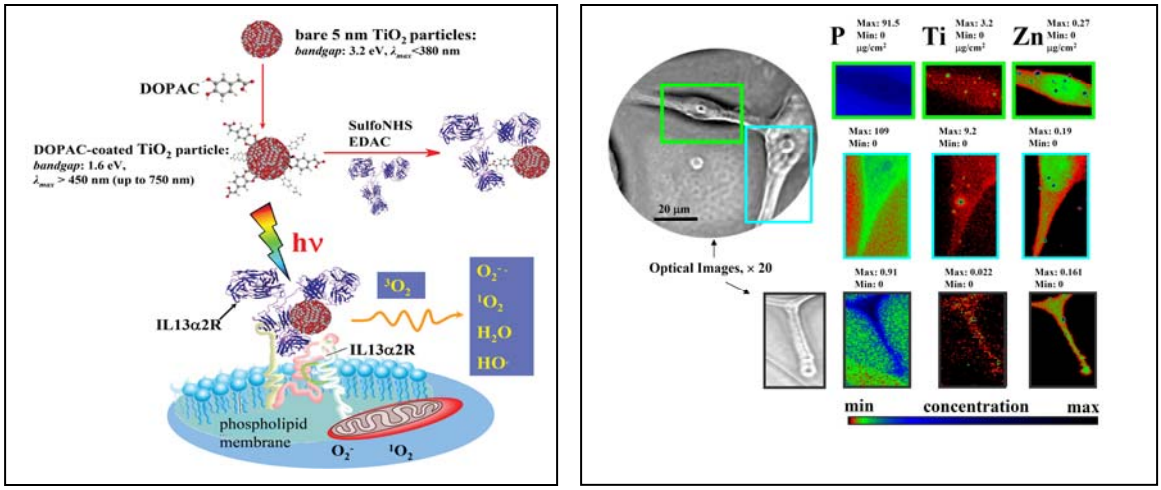

Figure1. General scheme
Nanobiocomposites consisted of 5 nm TiO_2 and IL13R recognizing antibody linked via DOPAC linker recognize and bind exclusively to surface IL13R. Visible light phoro-excitation of the nanobio hybrid in an aqueous solution results in formation of the various ROS. ROS, mainly superoxide cause cell membrane damage, permeability changes and cell death.

Figure2. X-ray fluorescence Microprobe-based visualization of the TiO_2-mAb binding to the single GBM cells (high antigen over-expressing A172 line). Elemental distribution of biogenic phosphorous and zinc are used to sketch cells and nucleus. Directed by the mAb exclusively to the surface of GB cells, TiO_2 nanoparticles are spread all through the GMB cell, including cells invadopodia (lower images). The intensity of the elemental images was displayed using a prism color table in logarithm scale which was shown to the bottom right. The max and minimum threshold values in micrograms per squared centimeter are given above each frame. Scans were obtained by using 10.0-keV incident energy with dwell times of 1 sec per pixel and 1-μm steps through the sample. Simultaneous appearance of intense "hot spots" in the Ti and Zn distribution images is possibly resulting of the titanium nanoparticles as well as other inorganic materials aggregation.

Future work: We will continue studies of TiO_2 nanobio hybrids interfacing with malignant mammalian cells using real-time PCR microarray analysis of the expression of key genes involved into ROS-based programmed cells death. Using novel advanced X-ray technique - Hard X-ray Nanoprobe we will study dynamics of important biochemical events following light-stimulus of photoreactive materials interfaced with mammalian cells. Furthermore, we will focus on development of other functional materials such as ferromagnetic particles with spin vortex ground state, for cellular signal transduction.

Publications:

N. M. Dimitrijevic, E. Rozhkova, T. Rajh, "Dynamics of Localized Charges in Dopamine-Modified TiO2 and their Effect on the Formation of Reactive Oxygen Species", *J. Am. Chem. Soc.*, 131, 2893-2899 (2009)

E. A. Rozhkova, V. Novosad, D.-H. Kim, J. Pearson, R. Divan, T. Rajh, and S. D. Bader "Ferromagnetic microdisks as carriers for biomedical applications" *J. Appl. Phys*., 105, 07B306 2009