ISSN: 1391-8796
Proceedings of 11th Ruhuna International Science & Technology Conference
University of Ruhuna, Matara, Sri Lanka
January 24, 2024

Keynote Speech

Tailored nanostructures for drug delivery and early cancer detection

Bossmann S.H., Covarrubias-Zambrano O., Neri-Sierra R., Kamath D., Payne M.

Department of Cancer Biology, The University of Kansas Medical Center, Kansas City, KS, U.S.A.

Abstract

The Bossmann group at the University of Kansas Medical Center is developing new nanotechnology to address key problems in cancer treatment. 1) With regard to drug delivery, we have developed poly(beta-amino ester)-based nanoparticles that use peptide signaling sequences to circumvent the reticuloendothelial system and to deliver their payload (small molecule drugs or mRNA) with precision to the sites of tumors and metastases. 2) To facilitate early detection of solid tumors, we have developed nanoparticle-based nanobiosensors for protease activities in serum since 2007. Recently, we have adapted a few-layer graphene as core nanostructure. This permits the design of long-term stable nanobiosensors capable of detecting pancreatic, lung, and ovarian cancer with very high precision at stage 1. The lower the cancer stage, the better are current treatment modalities.

Key words

Drug Delivery, Nanomedicine, Chimeric Signaling, Tumor Microenvironment, Graphene Nanobiosensor

Addressing the Challenge of Effective Drug Delivery

The discovery of effective Macromolecule Therapeutics has been significantly prolonged by the physiological differences between mice and (wo)men. Whereas a plethora of macromolecule/ nanodelivery systems exists to date that works very well in mouse models, there has been only limited success in humans [1, 2]. Therefore, “Systematically deliver macromolecules to intracellular targets for therapeutic benefit in cancer” has been declared a Cancer Grand Challenge by the National Cancer Institute (NCI) and Cancer Research UK (CRUK)[3]. We are addressing this challenge by chimeric signaling using attached signaling peptides. We are combining the “Don’t Eat Me Concept” in macromolecule delivery by CD-47 mimicry and by accelerating macromolecule uptake by tumors through the targeting of highly selective surface peptides.

The guiding paradigm of our research is that effective delivery of macro-molecule therapeutics to human tumors requires three consecutive steps. 1) Enhanced Extravasation, 2) Effective Targeting of Suitable Surface Receptors at the Surface of Cancer Cells, 3) Effective Uptake, Endosomal Escape, and Release of the Therapeutic Payload. Effective Macromolecule Therapeutics requires synergy of all three steps. A poly-beta-amino ester copolymer (PBAE) was selected as macromolecule for micro-RNA and drug delivery. The Bossmann group's versatile PBAE technology permits delivery of drugs to all cellular targets [4].

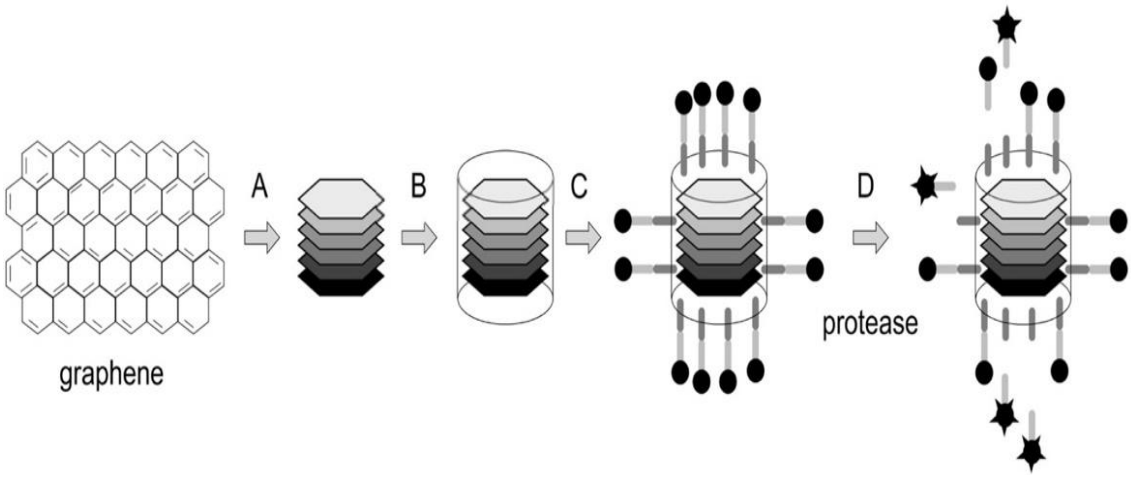
The Challenge of Early Cancer Diagnoses

Biomarkers[5] are biological molecules (enzymes, antibodies, peptides, etc.) that are present in bodily fluids or tissue and can be used for multiple diagnoses based on their expression and activity. The proposed study focuses on *proteases*[6], enzymes that degrade proteins. *Proteases* regulate signaling pathways and play essential roles in disease progression. Furthermore, the proteolytic network interacts with other important signaling pathways in cellular biology, involving chemokines, cytokines, and kinases[6]. Over 600 human proteases are active in regulating processes, approximately two percent of the human genome. For example, matrix metalloproteinases (MMPs) are a family of proteases that can degrade extracellular matrix (ECM), connective tissues, basement membranes, and have increased activity in inflammatory diseases including Cancer. One of the most important characteristics of proteases is that they form a proteolytic network and depend on activation by other proteases. This permits the selection of signature proteases at important nodes of this very complex network that are indicative as disease and/or inflammation markers. By utilizing multiple proteases as biomarkers, a crowd response can be obtained that is indicative of specific diseases or responses to interventions[7, 8].

Development of Graphene-based Nanobiosensors (G-NBS)

In 2020 Hawkeye Bio LLC, a development stage medical technology company based in Torrance, California, gained interest in this technology for lung cancer early detection. When initial collaboration work started, various challenges were faced with this NBS technology, which included poor long-term stability, expensive mass production due to the requirement of two fluorescent dyes (one for Förster Resonance Energy Transfer (FRET) quenching and one for sensing), relatively poor repeatability, and Fe/Fe_3O_4 nanoparticles becoming magnetized during shipping, which significantly reduced its water dispersion properties. For this reason, a newer novel NBS was developed, replacing the Fe/Fe_3O_4 core with detonation graphene. The new derived graphene-NBS has overcome the major challenges faced by the previous Fe/Fe_3O_4 generation. graphene-NBSs are highly water/buffer dispersible, and they have a superior long-term stability.

Explosion graphene's optical absorption properties makes it a very effective quencher for fluorescent molecules. Therefore, only one dye is required for the new novel technology.



Design of NBS (A) Few-layer graphene (n=5-7); (B) Addition of the TEG4amine layer; (C) Anchoring of the consensus sequences + fluorescent dye (tetrakis-carboxycarbonyl-porphyrin, TCPP) via amide bonds; (D) proteolytic activation: The consensus sequences is cleaved, and the fluorescence of the dye is activated.

Graphene-NBS consists of a series of reactions to modify the surface layer of graphene. First, bromovaleric acid is added to explosion graphene to form a carboxygraphene (CG) layered intermediate, which is then further coated with branched polyethylenimine (PEI) to generate G-PEI intermediate, and finally, a TCPP-labeled oligopeptide is linked to complete the graphene-PEI NBS.

To further improve this technology, Covarrubias and Bossmann have developed a newer Generation 2.0 of graphene-NBS, saving steps to modify the surface layer of graphene, which will be the focus of my presentation.

Acknowledgements

Dr. Stefan H. Bossmann acknowledges the intellectual contributions from Drs. Thilani Samarakoon, Dinusha Udukala, Harshi Manawadu, and Madumali Kalubowilage, as well as financial support from NSF (EFRI 2129617) and NIH (R01 CA284065-01)

References:

1. Bertrand N, Wu J, Xu X, Kamaly N, Farokhzad OC. Cancer nanotechnology: the impact of passive and active targeting in the era of modern cancer biology. Advanced drug delivery reviews. 2014;66:2-25. Epub 2013/11/22. doi: 10.1016/j.addr.2013.11.009. PubMed PMID: 24270007.

2. Shi J, Kantoff PW, Wooster R, Farokhzad OC. Cancer nanomedicine: progress, challenges and opportunities. Nature reviews Cancer. 2017;17(1):20-37. Epub 2016/11/11. doi: 10.1038/nrc.2016.108. PubMed PMID: 27834398.

3. https://www.cancer.gov/grants-training/grants-funding/cancer-grand-challenges.

4. Covarrubias-Zambrano O, Shrestha TB, Pyle M, Montes-Gonzalez M, Troyer DL, Bossmann SH. Development of a Gene Delivery System Composed of a Cell-Penetrating Peptide and a Nontoxic Polymer. ACS Appl Bio Mater. 2020;3(11):7418-27. doi: 10.1021/acsabm.0c00561.

5. Aronson JK, Ferner RE. Biomarkers-A General Review. Curr Protoc Pharmacol. 2017;76:9.23.1-9..17. Epub 20170317. doi: 10.1002/cpph.19. PubMed PMID: 28306150.

6. Mason SD, Joyce JA. Proteolytic networks in cancer. Trends Cell Biol. 2011;21(4):228-37. doi: 10.1016/j.tcb.2010.12.002. PubMed PMID: WOS:000290054000005.

7. Kasi A, Sun W, Ehsan S, Covarrubias J, Eschliman K, Neri R, Agarwal D, Zambrano OC, Bossmann SH, Natarajan B. Early detection of pancreatic cancers by novel nanobiosensor-based protease biomarkers using hierarchical decision structure. Journal of Clinical Oncology. 2021;39(15_suppl):e16273-e. doi: 10.1200/JCO.2021.39.15_suppl.e16273.

8. Covarrubias-Zambrano O, Agarwal D, Kalubowilage M, Ehsan S, Yapa AS, Covarrubias J, Kasi A, Natarajan B, Bossmann SH. A New Hope for Liquid Biopsies: Early Detection of Pancreatic Cancer By Means of Protease Activity Detection in Serum Applying a Hierarchical Decision Structure. medRxiv. 2022:2022.10.18.22281240. doi: 10.1101/2022.10.18.22281240.