ELSEVIER

Nanomedicine: Nanotechnology, Biology, and Medicine
xx (2015) xxx – xxx

nanomedicine
Nanotechnology, Biology, and Medicine

nanomedjournal.com

Biological interactions of carbon-based nanomaterials: From coronation to degradation

Kunal Bhattacharya, PhD[a], Sourav P. Mukherjee, PhD[a], Audrey Gallud, PhD[a], Seth C. Burkert, MSc[b], Silvia Bistarelli, MSc[c], Stefano Bellucci, PhD[c], Massimo Bottini, PhD[d,e], Alexander Star, PhD[b], Bengt Fadeel, MD, PhD[a,*]

[a]*Division of Molecular Toxicology, Institute of Environmental Medicine, Karolinska Institutet, Stockholm, Sweden*
[b]*Department of Chemistry, University of Pittsburgh, Pittsburgh, PA, USA*
[c]*National Institute of Nuclear Physics–INFN, Frascati, Province of Rome, Italy*
[d]*Department of Experimental Medicine and Surgery, University of Rome 'Tor Vergata', Rome, Italy*
[e]*Sanford Burnham Prebys Medical Discovery Institute, La Jolla, CA, United States*

Received 19 August 2015; accepted 10 November 2015

Abstract

Carbon-based nanomaterials including carbon nanotubes, graphene oxide, fullerenes and nanodiamonds are potential candidates for various applications in medicine such as drug delivery and imaging. However, the successful translation of nanomaterials for biomedical applications is predicated on a detailed understanding of the biological interactions of these materials. Indeed, the potential impact of the so-called bio-corona of proteins, lipids, and other biomolecules on the fate of nanomaterials in the body should not be ignored. Enzymatic degradation of carbon-based nanomaterials by immune-competent cells serves as a special case of bio-corona interactions with important implications for the medical use of such nanomaterials. In the present review, we highlight emerging biomedical applications of carbon-based nanomaterials. We also discuss recent studies on nanomaterial 'coronation' and how this impacts on biodistribution and targeting along with studies on the enzymatic degradation of carbon-based nanomaterials, and the role of surface modification of nanomaterials for these biological interactions.

Key words: Carbon nanotubes; Graphene oxide; Fullerenes; Nanodiamonds; Biodegradation; Bio-corona

Engineered nanomaterials provide unique advantages and opportunities in several areas of medicine including therapeutics, diagnostics, imaging, and regenerative medicine.[1,2] Carbon-based nanomaterials such as fullerenes, carbon nanotubes, carbon nanohorns, carbon nanodots, nanodiamonds, and graphene and its derivatives have unique electronic, optical, thermal, and mechanical properties and have attracted considerable attention in recent years in nanomedicine.[3-5] Hence, many studies have attempted to exploit these materials for drug delivery or imaging, or both. As pointed out in a recent editorial, the successful commercialization of nanomedicines ultimately depends on demonstrating their superiority over existing approaches and on documenting their safety.[2] Indeed, a detailed understanding of the biological interactions of nanomaterials, not least the interactions with cellular and other components of the immune system (Figure 1) is important both from an efficacy

Funding: This work was supported by grants from the European Commission (Flagship Project GRAPHENE, grant no. 604391; FP7-NANOSOLUTIONS, grant no. 309329; FP7-NANOREG, grant no. 310584), the Swedish Research Council, and the Swedish Research Council for Environment, Agricultural Sciences and Spatial Planning (FORMAS) (to B.F.); National Institute of Environmental Health Sciences (NIEHS) grant no. R01ES019304 (to A.S.); Arthritis National Research Foundation (John Vaughan Scholarship) (to M.B.); and SfP NATO grant no. SfP-984537, and Italian Ministry of Health grant no. PE-2011-02347026 (to S.B.).

Conflict of interest: The authors declare no conflicts of interest.

Portions of the graphical abstract were reproduced with permission from Nature Publishing Group and John Wiley & Sons.

Acknowledgements.: We thank Dr. Kjell Hultenby, Karolinska Institutet, for assistance with SEM.

*Corresponding author at: Institute of Environmental Medicine, Karolinska Institutet, Stockholm, Sweden.

E-mail address: bengt.fadeel@ki.se (B. Fadeel).

http://dx.doi.org/10.1016/j.nano.2015.11.011

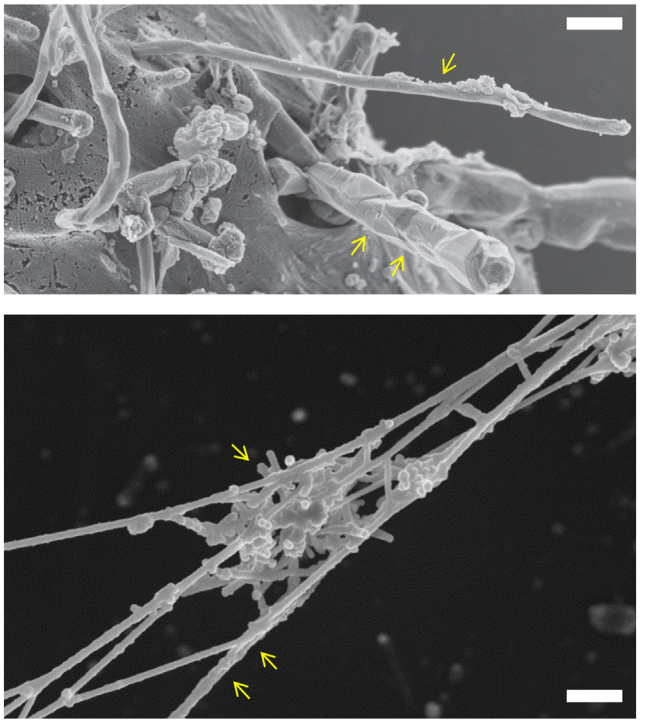

Figure 1. Cellular and extracellular interactions of carbon nanotubes. The upper panel shows an SEM image of isolated MWCNTs (single arrow) or a bundle of MWCNTs (two arrows) entering human mesothelial cells. Reprinted from: Shi X, von dem Bussche A, Hurt RH, Kane AB, Gao H. Cell entry of one-dimensional nanomaterials occurs by tip recognition and rotation. Nat Nanotechnol. 2011;6(11):714-9, with permission from Nature Publishing Group. The lower panel shows a cluster of short-cut SWCNTs (single arrow) entrapped in chromatin fibers (two arrows) of purified neutrophil extracellular traps [see Farrera et al[6] for further details]. SEM courtesy of K. Hultenby, Karolinska Institutet.

and safety point of view, as is the understanding of the ultimate fate of the nanomaterial – accumulation, degradation, and/or excretion – in the human body.[7] To this end, particular attention should be devoted to the role of adsorbed biomolecules which may confer a new biological 'identity' to nanomaterials,[8] and is likely to play an important role for cellular uptake and *in vivo* biodistribution of nanomaterials.[9]

Detailed accounts of the routes of synthesis and the physicochemical properties of carbon-based nanomaterials are beyond the scope of the present review, but a brief introduction is provided here. Fullerenes are entirely composed of carbon and have the form of spheres, ellipsoids or tubules. Spherical and cylindrical fullerenes are also referred to as buckyballs and buckytubes (or carbon nanotubes), respectively. The first representative of the buckyball family, referred to as buckminsterfullerene, is composed of 60 carbon atoms (C_{60}) and has the shape of a truncated icosahedron with 20 hexagons and 12 pentagons and a diameter of approximately 1 nm, thus resembling a football (in the United States, a soccer ball); indeed, a picture of a football was included in the very first publication, and the authors even contemplated the alternative name, soccerene.[10] Iijima is credited with the discovery of carbon nanotubes (CNTs)[11] although some claim that these structures ("graphitic carbon needles") had been observed decades earlier.[12] CNTs are graphitic tubules, which can be capped with hemifullerenes at the ends, consisting of a single graphene sheet (single-walled carbon nanotubes, SWCNTs) or several concentric and nested sheets (multi-walled carbon nanotubes, MWCNTs). Both types of CNTs have nano-scale dimensions and display a very high aspect ratio, i.e., the ratio between the length and the diameter of the material. Hence, SWCNTs have a diameter of approximately 1 nm and lengths up to a few microns or more, whereas MWCNTs have diameters of several tens of nanometers and lengths up to several tens of microns or more. All of the aforementioned nanomaterials can be related to a parent material known as graphene consisting of a single atomically thin sheet of hexagonally bound sp^2 carbon atoms.[13] For a comprehensive overview of the structural, electronic, and biological properties and applications of graphene and other 2-D materials, see Ferrari et al.[14] Nanodiamonds represent yet another class of nanoparticles in the carbon family, with highly versatile physical and chemical properties.[15] They are mainly composed of carbon sp^3 structures in the core, with sp^2 and disorder/defect carbons on the surface, and display single-digit nm sizes.

In the present review, we will highlight emerging biomedical applications of various carbon-based nanomaterials. We will also discuss bio-corona formation and the propensity for enzymatic degradation, especially with regards to CNTs and graphene oxide (GO), which are the most intensively investigated carbon-based nanomaterials to date in the field of nanomedicine, along with fullerenes and nanodiamonds. The impact of surface modifications, including grafting of polymers, on the biological interactions of these materials is also highlighted.

Biocompatibility of carbon-based nanomaterials

Being small confers advantages in terms of negotiating biological barriers, which may be desirable, but nanoscale size *per se* is not sufficient to qualify as a nanotechnology.[16] Carbon-based nanomaterials, however, possess intrinsic physicochemical properties that can potentially be exploited. For instance, CNTs display strong optical absorption in the near infrared, Raman scattering as well as photo-acoustic properties that widen the scope of *in vivo* applications as they can potentially have bio-imaging and tracing functions coupled with drug delivery.[4] Graphene is another material with many promising areas of application as a result of its large surface area and possibility of easy functionalization, providing opportunities for drug delivery.[5] Moreover, its unique mechanical properties suggest tissue engineering and regenerative medicine applications.[17] Other carbon-based nanomaterials such as fullerenes and nanodiamonds have also received much attention in recent years, with emphasis mainly in the area of cancer medicine.[4] In the present review, we will highlight some illustrative, pre-clinical examples from recent literature.

However, safety first. The potential toxicity of carbon-based nanomaterials has been the subject of much concern in the past decade and much skepticism initially surrounded the notion of using, in particular, CNTs as drug delivery systems due to the fact that these fiber-like materials were presumed to be biopersistent, and, therefore, to possess asbestos-like pathogenicity.[18-20]