

Communication

Handling and Risk Mitigation of Nanoscale Graphene and Related Materials: Some Considerations and Recommendations

Velram Balaji Mohan 

Centre for Advanced Composite Materials and Plastics Centre of Excellence, Department of Mechanical Engineering, the University of Auckland, Khyber Pass Road, Newmarket, Auckland 314-390, New Zealand; vmoh005@aucklanduni.ac.nz; Tel.: +64-9-923-3681

Received: 14 May 2019; Accepted: 24 June 2019; Published: 1 July 2019



Abstract: The purpose of this communication is to put forward some considerations and recommendations while handling nanomaterials, especially graphene and its derivatives. A large graphene sheet is generally stable and inert; thus, graphene and its derivatives are not considered hazardous, but good laboratory practices should be taken seriously for the safe handling and use of such materials. This article provides some insights about nanoscale graphene handling and some important considerations.

Keywords: graphene; carbon; nanoparticle; handling; safety; precautions

1. Introduction

Graphene platelets are best described as a nanomaterial, consistent with international standards. ISO/TS 80004-1:2010 defines a nanomaterial as a “material that has dimensions in the nanoscale (typically 1–100 nm), either externally or internally” [1].

In 2011, European Commission (EC) published a definition of a nanomaterial and recommended Registration, Evaluation, Authorisation and Restriction of Chemicals (REACH) and Classification, Labelling and Packaging (CLP) to adapt with the definition under (2011/696/EU). Based on this recommendation, a nanomaterial refers to any materials (organic, inorganic in the form of unbound or agglomerated state with the size between 1–100 nm. Figure 1 shows the classification of grapheneic materials and scanning electron microscope images of nanographene.

Handling and disposal of nanomaterials is an important ongoing topic, and there needs to be a constant discussion about it to avoid any potential risk of respiratory hazards. While there are studies focused on safety considerations and precautions, regular update on such subjects should be made available due to increased use of graphene nanomaterials. This article refers to some of the most reliable literature and experimental studies on safety, handling, risk mitigation and adverse effect of nanomaterials, and attempts (in a concise manner) to make some important considerations and precautionary measures for readers.

It is important to address the safety concerns of nanomaterial and propose possible aids to avoid risks, as graphene and their derivatives have become promising candidates in wide range of applications such as flexible devices, electrodes, sensors, composites as performance-enhancing fillers, biomedical, conductive inks, thin conductive films, drug delivery, micro-robotics, optoelectronics, energy storage devices, nano-filters, coatings, membranes, photovoltaic cells and bionic devices [2–7].

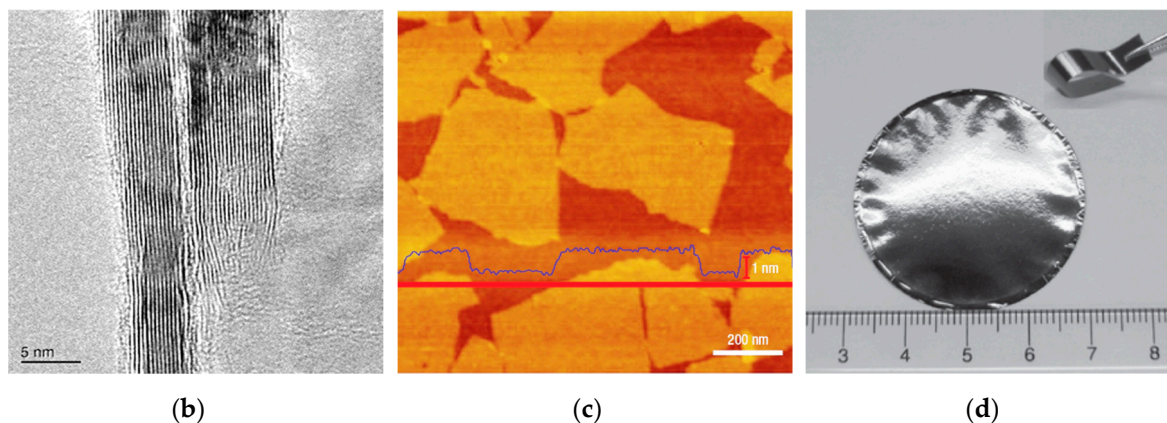
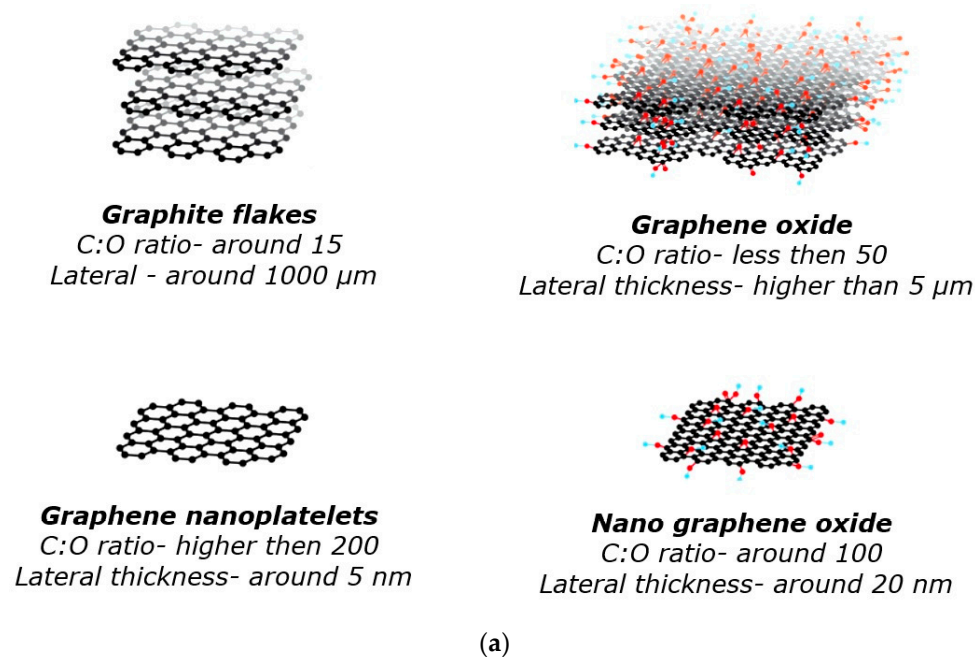


Figure 1. (a) Major classification of graphitic materials, (b) TEM image of graphene nanoplatelets GNP [8], (c) AFM image of GNP suspension on a substrate [9] and (d) chemically converted graphene film reproduced with permission from [9]. Springer Nature, 2008

2. Materials and Methods

Graphene nanoplatelets used in this study were purchased from Scharlau and Emfutur Tech (called as GNS 1 in this work), Spain Easchem Co., Ltd., Changsha, China (called as GNS 2 in this work).

Particle size analysis was done by laser diffraction with the particle size analyser HELOS from Sympatec GmbH (Clausthal-Zellerfeld, Germany), in combination with the dry dispersing unit RODOS and with the micro-dosing unit ASPIROS. Figure 2 shows the particle size distribution of the GNPs.

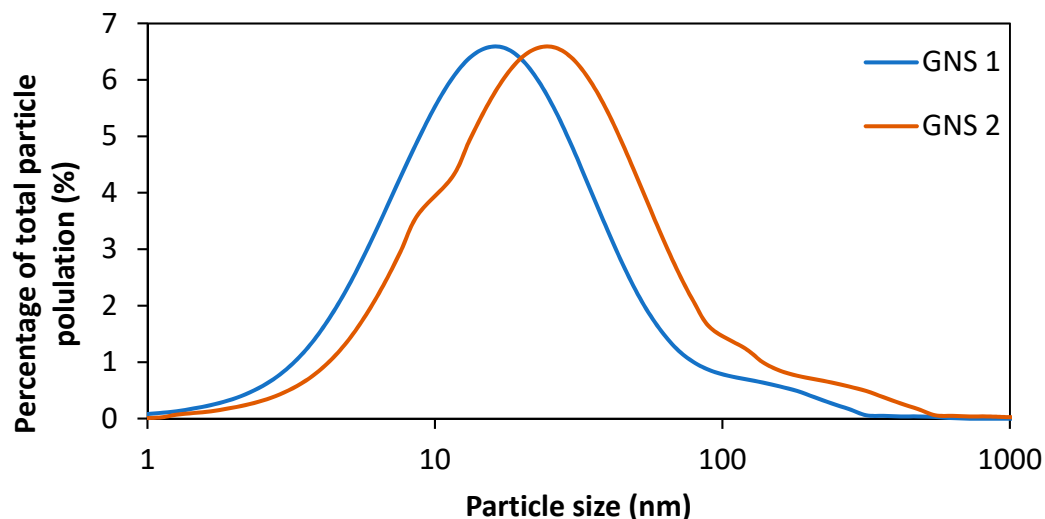


Figure 2. Particle size distribution for two different GNPs supplied by different vendors.

Nomenclature

The following nomenclature has been used to classify different graphene and other materials:

1. Graphene as a generic term for nanoscale graphene
2. Graphene nanoplatelets (GNP)
3. Graphene nanosheets (GNS)
4. Graphene oxide (GO)
5. Reduced graphene oxide (rGO)
6. Functionalised graphene (fG)
7. Carbon nanotube (CNT)

3. Respiratory Hazards Associated with Graphene Nanoplatelets (GNP)

Graphene platelets (GNP) have aerodynamic properties (in particular aerodynamic diameter or *D_{ae}*) far lower than their projected area would suggest [10]. GNP with areas ranging between 5–30 micrometres have an aerodynamic diameter between 1 and 3 micrometres and will fall within respirable size fraction that will deposit beyond the ciliated airways.

Using animal models exposed to GNP, Schinwald et al. [10] found an inflammatory response and macrophage recruitment both in lung and pleural space consistent with ‘frustrated phagocytosis’. This inflammatory response waned over time, and there was a clearance of the graphene to the lymph nodes suggesting that the GNP were gradually broken up over time. However, a significant fraction of the GNPs do remain in the pleural cavity. These studies in animal models did not examine prolonged exposure, but prolonged chronic inflammation that would result from prolonged exposure is highly likely to damage lung tissue permanently. The above strongly indicates a respiratory hazard, which has been documented and has been corroborated in other testing regimes [11].

Pathology would be expected to follow the fibre paradigm, due to the fact that GNP sheets cannot be removed efficiently because their flakes are not efficiently cleared because of their rigidity, insolubility, dimensions and subsequently frustrated phagocytosis [10–12]. Any risk assessment needs not only to take into account the respiratory hazard, but the elevated risk pathology of sensitive groups such as asthma sufferers. It is important to stress that the toxicity of the graphene materials should not be compared with that of CNTs as they behave differently (though they affect the biological systems by the size of tubes and chemical entities they carry along) with the biological system as they come in to contact with them. In addition, the mechanism of interaction by CNTs is harmful due to their structural (cylindrical) and dimensional nature. This is not the case with graphene derivatives as

they are in sheet form mostly [13]. However, CNTs in ribbon form can also be carcinogenic due to their fibrous architecture. It is worth mentioning that the environmental effects and behaviour of engineering nanomaterials (ENM) should be extensively studied as there is very little knowledge available. A short review based on case studies of the toxicity of CNT was reported by Nowack et al. [14] in which it has been said that the release of CNT in the environment during production and application can have serious effects on human systems due to their carcinogenicity.

Furthermore, incineration and landfill disposal of CNT can also have airborne contamination, and proper protocols should be developed to understand the role of toxic responses of ENM to the environments as well. The toxicity/carcinogenicity of MWCNT has been found to affect lung fibroblasts via collagen production (Stanton hypothesis), which has been reported by He et al. [15]. The study also indicated that the high-level formation of collagen triggered pulmonary fibrosis in human systems.

A commentator on Donaldson's work has raised the issue of whether GNPs/GNSs could, in fact, become airborne [16]. An evaluation survey of a manufacturing and refining facility with offices and research laboratories indicates that GNPs/GNSs do become airborne [17], while no noticeable particle emissions occurred when fume hoods and glove boxes were used as control measures for research laboratory activities, indicating that conventional local ventilation measures in laboratories are effective. However, high particle concentrations were found in the office and laboratories as a result of spillover from production facilities where air extraction was found to be inadequate to prevent airborne contamination. Separating ventilation systems for production and non-production areas, and providing a positive pressure for office areas, would prevent particles being entrained into non-production areas.

Furthermore, the following finding on graphene interaction with biological systems from different sources have been summarised by Park et al. [12]:

- Graphene nanoparticles with a high specific surface area at the nanoscale can have higher reactivity;
- Graphene-based aerosols can penetrate at a higher rate when their mass median aerodynamic diameter is $<3\ \mu\text{m}$;
- Graphene of size $<1\ \mu\text{m}$ was found to be going through lungs less than that of smaller sized materials;
- GO sheets of larger size were assessed to be having a stronger antibacterial activity to *Escherichia coli* (*E-coli*) than those of smaller sheets;
- Cell membranes can easily be affected by sharp edges of the sheets;
- The lung macrophages were less susceptible to when the aspect ratio of nanomaterials are >3 and a length of at least $10\ \mu\text{m}$;
- Increased agglomeration/aggregation of GNP can reduce the hemolytic activity;
- Aggregations/agglomerate can be formed by high concentrations of nanoparticles, by which translocation may be reduced in biological blockades;
- Human neutrophil based *myeloperoxidase* can degrade highly dispersed graphene nanoparticles, while the agglomerated state of nanoparticles does not get degraded by the same (Figure 3 shows the effect of different graphene materials on human organs upon exposure); however, the degradation of GNP may not be directly related to the organs where the GNP precipitate;
- Hydrophilic graphene is found to be more biocompatible than hydrophobic graphene;
- Biological membranes, such as Gram-positive versus Gram-negative bacteria can be influenced by altered surface charges;
- Surface contaminants of graphene may cause potential harm to the human system.

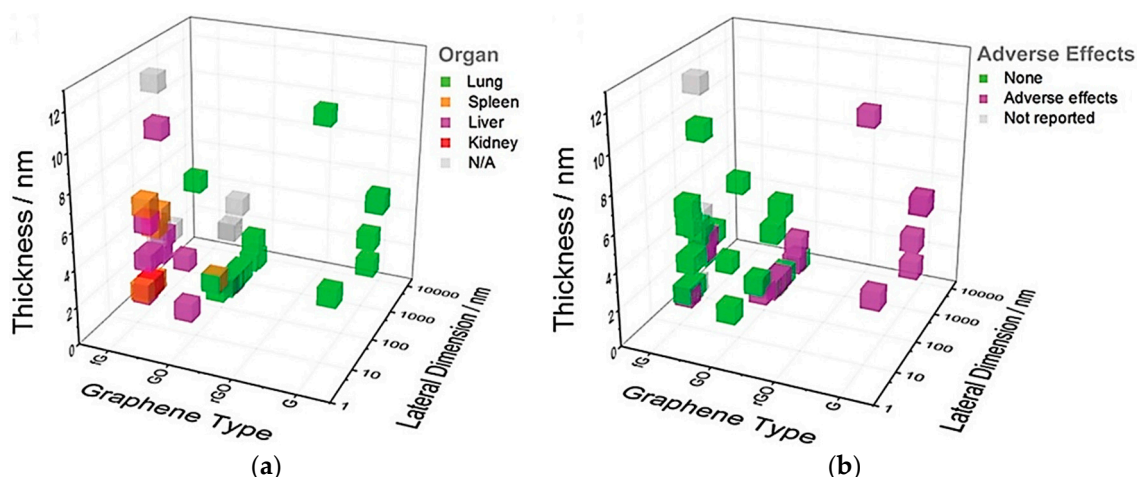


Figure 3. Graphical representation of (a) effect of graphene aggregates on human organs and (b) negative effects reported in earlier studies, reproduced with permission from [18]. Royal Society of Chemistry, 2015.

4. Considerations and Recommendations

With the use of proper lab and work practice, engineering controls and appropriate equipment and aids, it is highly possible to reduce and mitigate the risk associated with nanomaterial handling. The following are some important considerations for nanomaterial handling and disposal:

1. Given that the extent of any potential airborne particle contamination is unknown, it is recommended that a particle survey should be undertaken to quantify any airborne particle contamination with graphene. If airborne contamination is detected, such a survey would help with risk mitigation measures such as the provision of local, flexible enclosures, or rebalancing local ventilation systems in a laboratory.
2. Identification of processes and procedures in which graphene is used, and where it could become airborne. It would help in the understanding of how local ventilation systems are used in each of these processes, and inform how risk mitigation measures could be improved. Possible exposure monitoring devices would be helpful to identify ENM contamination. Strategic goals should be developed in order to protect researchers using ENM and to translate the research findings of the effects of ENM into workplace practice efficiently [19].
3. Gaining confirmation from manufactures of P3 masks would provide adequate protection for graphene with a Dae of 3 microns and, if necessary, ensure the masks are individually fitted.
4. To reduce the risk of handling graphene:
 - Limiting the amount of nanomaterials or batch size of samples would reduce the risk of airborne contamination and exposure to the skin [20];
 - Containing graphene in a closed cabinet surrounded by appropriate materials is necessary to avoid spills [21];
 - When preparing dispersions, contain graphene as much as possible, particularly while it is being subjected to ultrasonication;
 - It is also wise to inform co-workers/researchers who are not working with ENM to alert them how to mitigate risks associated with such materials;
 - Safe-by-design tactics changing the biological activity via reducing the toxicity-related effects will have high effectiveness in occupational risk control hierarchy, which would highly suitable for ENM.
5. Reviewing of clean up and waste disposal procedures is necessary to ensure these are adequate and up to date. Vacuum cleaners fitted with high efficiency particulate arrestance (HEPA) filters

should be used. Waste containing or contaminated with graphene should be separately bagged and sent out for deep burial by authorised personnel.

6. A fume hood with HEPA filters and a weighing balance should be dedicated for graphene to avoid any sort of contamination. All of the weighing processes are carried out under the fume hood. Likely consequences of exposure to graphene particles over 2 μm are respiratory or skin irritation/inflammation.
7. Use a vacuum cleaner with a HEPA filter for loose powder:
 - Small powder spills (typically involving less than 5 mg of material) of nano or micron-sized graphene must be removed using cloth/gauze/absorbent using appropriate removal material (for example, soapy water). The contaminated surfaces must be cleaned several times until no trace of nanomaterial is visible. One can use a clear cloth to make the surface is cleaned enough. The contaminated clean-up materials should be disposed of in an appropriate way [11,20,22];
 - Large spills need to be taken care of by professionally trained cleaners;
 - Figure 4 shows nano-enclosure and protective measures while handling ENM.
8. Facilities handling nanomaterials should have a clear and concise safe work instruction (SWI) manual, log sheet and drill press document to allow users (particularly new users) to be aware of safe handling and housekeeping.
9. A proper and detailed risk assessment should be carried out for any class of nanomaterials that is under usage or experimentation. This will inform the technician and users on how to mitigate the problem when it arises. The document should be regularly updated and revised upon necessity [23].
10. Disposal of nanoparticles [24]:
 - Graphene materials should not be disposed of as general waste compounds in sinks or drains;
 - If any ENM is dispersed in any acid medium or reductants, it should be neutralised before disposal;
 - Decanting liquids containing graphene should be disposed of with the help of chemical waste disposal companies;
 - The appropriate disposal protocols should be sufficiently communicated to all the users and non-users, and integration between different scientific disciplines should be properly carried out.

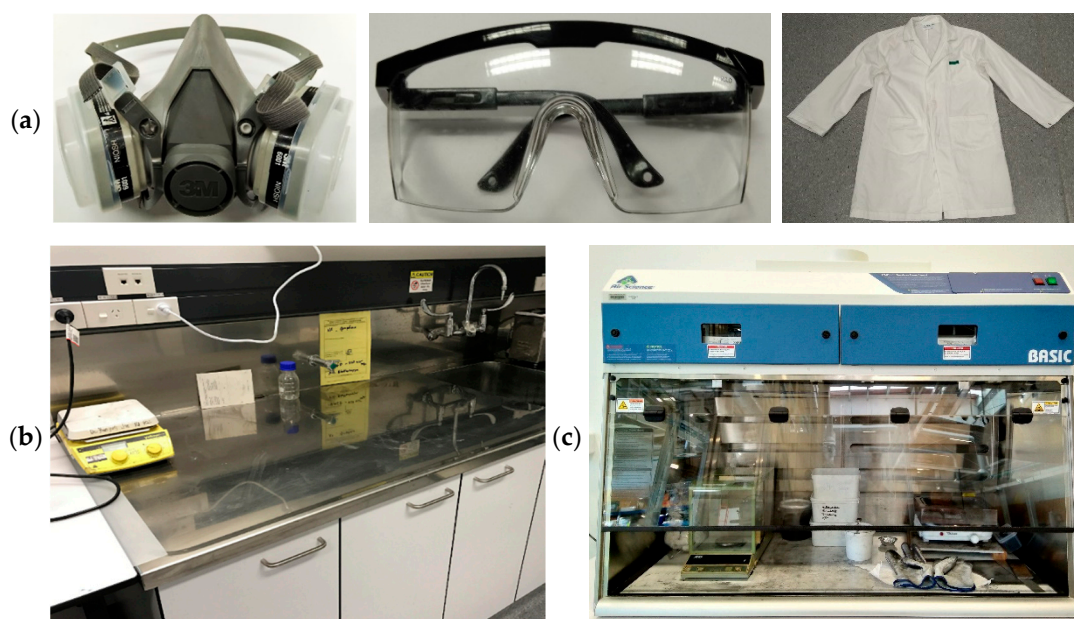


Figure 4. (a) Illustration of basic personal protective wear while handling with nanomaterials under nano-enclosure (HEPA filtered mask, fully covered eye glasses and lab coat), (b) image of after cleaning of working areas, and (c) a typical nano-enclosure equipped with HEPA filter where graphene or similar nanomaterials should be handled under.

5. Summary

The available knowledge on nanomaterials' handling, disposal, toxicity and their interaction with biological systems are still inadequate. However, to be able to have risk assessments, one has to read a sufficient amount of literature and have safety considerations accessible. As of now, there is no equipment that evaluates or details routine exposure measurement and threshold limit values of nanomaterials. However, based on the literature study, considerable evaluations have been made to understand the mechanism of interaction of nanomaterials with biological systems, and it has been shown that they do have negative effects. Hence, all possible precautions should be made available, and extensive risk assessment/mitigation protocols should be developed and used. However, a generalised approach for a different state, such as powder, dispersion, suspension or in polymer blends cannot be maintained; thus, knowledge of handling different states of nanomaterials is required. Research facilities/laboratories should have pragmatic mitigation procedures to control the exposure and airborne contamination of nanomaterials, as well as for specific activities involving nanomaterials. It is important to note that the considerations and suggestions made in this article are **provisional** with the available knowledge, and in future reliable assessment and scientific data should be considered. Furthermore, considering that safety risks will be entirely dependent on the specific properties, characteristics, and use of the material in each study, generalisation about the toxicity profile of genres of materials needs to be avoided [20]. In addition, proper communication between the manufacturer, supplier and end-user about the nanomaterials' functionality and safety should be done.

Funding: This research received no external funding.

Conflicts of Interest: The author declares no conflicts of interest.

References and Notes

1. *Code of Federal Regulations: Title 40—Protection of Environment: Chapter 1—Environmental Protection Agency (Continued). Subchapter R-Toxic Substances Control Act. Part 721—Significant New Uses of Chemical Substances;* Environmental Protection Agency: Washington, DC, USA, 2005.

2. Kurian, A.S.; Mohan, V.B.; Bhattacharyya, D. Embedded large strain sensors with graphene-carbon black-silicone rubber composites. *Sens. Actuators A: Phys.* **2018**, *282*, 206–214. [CrossRef]
3. Mohan, V.B.; Brown, R.; Jayaraman, K.; Bhattacharyya, D. Optimisation of hybridisation effect in graphene reinforced polymer nanocomposites. *Adv. Compos. Mater.* **2017**, 1–17. [CrossRef]
4. Mohan, V.B.; Jayaraman, K.; Bhattacharyya, D. Relevance of adhesion in particulate/fibre- polymer composites and particle coated fibre yarns: A critical review. *Rev. Adhes. Adhes.* **2016**, *4*, 119–151. [CrossRef]
5. Mohan, V.B.; Jayaraman, K.; Bhattacharyya, D. Fabrication of highly conductive graphene particle-coated fiber yarns using polymeric binders through efficient coating techniques. *Adv. Polym. Technol.* **2018**. [CrossRef]
6. Mohan, V.B.; Krebs, B.J.; Bhattacharyya, D. Development of novel highly conductive 3D printable hybrid polymer-graphene composites. *Mater. Today Commun.* **2018**, *17*, 554–561. [CrossRef]
7. Mohan, V.B.; Lau, K.-T.; Hui, D.; Bhattacharyya, D. Graphene-based materials and their composites: A review on production, applications and product limitations. *Compos. Part B: Eng.* **2018**, *142*, 200–220. [CrossRef]
8. SEM of graphene nanoplatelets—Supplied by Emfutur Technologies Spain.
9. Li, D.; Müller, M.B.; Gilje, S.; Kaner, R.B.; Wallace, G.G. Processable aqueous dispersions of graphene nanosheets. *Nat. Nanotechnol.* **2008**, *3*, 101. [CrossRef] [PubMed]
10. Schinwald, A.; Murphy, F.A.; Jones, A.; MacNee, W.; Donaldson, K. Graphene-based nanoplatelets: A new risk to the respiratory system as a consequence of their unusual aerodynamic properties. *ACS Nano* **2012**, *6*, 736–746. [CrossRef] [PubMed]
11. Bianco, A. Graphene: Safe or toxic? The two faces of the medal. *Angew. Chem. Int. Ed.* **2013**, *52*, 4986–4997. [CrossRef] [PubMed]
12. Park, M.V.; Bleeker, E.A.; Brand, W.; Cassee, F.R.; van Elk, M.; Gosens, I.; de Jong, W.H.; Meesters, J.A.; Peijnenburg, W.J.; Quik, J.T. Considerations for safe innovation: The case of Graphene. *ACS Nano* **2017**, *11*, 9574–9593. [CrossRef] [PubMed]
13. Graphene Toxicity—The Interview by The Graphene Council dated July 2014.
14. Nowack, B.; Ranville, J.F.; Diamond, S.; Gallego-Urrea, J.A.; Metcalfe, C.; Rose, J.; Horne, N.; Koelmans, A.A.; Klaine, S.J. Potential scenarios for nanomaterial release and subsequent alteration in the environment. *Environ. Toxicol. Chem.* **2012**, *31*, 50–59. [CrossRef] [PubMed]
15. He, X.; Kiratipaiboon, C.; Porter, D.W.; Rojanasakul, L.W.; Dinu, C.Z.; Wang, K.; Yang, Y.; Rojanasakul, Y. Predicting Nanotube Fibrogenicity through Stem Cell-Mediated Fibroblast Focus and Spheroid Formation. *Nano Lett.* **2018**, *18*, 6500–6508. [CrossRef] [PubMed]
16. Bradley, D. Is graphene safe? *Mater. Today* **2012**, *15*, 230. [CrossRef]
17. Lo, L.M.; Hammond, D.; Bartholomew, I.; Almaguer, D.; Heitbrink, W.; Topmiller, J. *Engineering Controls for Nano-Scale Graphene Platelets During Manufacturing and Handling Processes*; Dep. Health Hum. Serv. Cent. Dis. Control. Prev. Natl Inst Occup. Safety Health: Washington, DC, USA, 2011.
18. Bussy, C.; Jasim, D.; Lozano, N.; Terry, D.; Kostarelos, K. The current graphene safety landscape—A literature mining exercise. *Nanoscale* **2015**, *7*, 6432–6435. [CrossRef] [PubMed]
19. Kuempel, E.D.; Geraci, C.L.; Schulte, P.A. Risk assessment and risk management of nanomaterials in the workplace: Translating research to practice. *Ann. Occup. Hyg.* **2012**, *56*, 491–505. [PubMed]
20. Bussy, C.; Ali-Boucetta, H.; Kostarelos, K. Safety considerations for graphene: Lessons learnt from carbon nanotubes. *Acc. Chem. Res.* **2012**, *46*, 692–701. [CrossRef] [PubMed]
21. Laux, P.; Tentschert, J.; Riebeling, C.; Braeuning, A.; Creutzenberg, O.; Epp, A.; Fessard, V.; Haas, K.H.; Haase, A.; Hund-Rinke, K. Nanomaterials: Certain aspects of application, risk assessment and risk communication. *Arch. Toxicol.* **2018**, *92*, 121–141. [CrossRef] [PubMed]
22. Freeland, J.; Hulme, J.; Kinnison, D.; Mitchell, A.; Veitch, P.; Aitken, R.; Hankin, S.; Poland, C.; Bard, D.; Gibson, R.; et al. Working Safely with Nanomaterials in Research & Development by The UK NanoSafety Partnership Group (UKNSPG). Available online: <https://www.safenano.org/media/64896/Working%20Safely%20with%20Nanomaterials%20-%20Release%201%2000%20-%20Aug2012.pdf> (accessed on 15 May 2019).

23. Seabra, A.B.; Paula, A.J.; de Lima, R.; Alves, O.L.; Durán, N. Nanotoxicity of graphene and graphene oxide. *Chem. Res. Toxicol.* **2014**, *27*, 159–168. [[CrossRef](#)] [[PubMed](#)]
24. Kong, B.; Seog, J.H.; Graham, L.M.; Lee, S.B. Experimental considerations on the cytotoxicity of nanoparticles. *Nanomedicine* **2011**, *6*, 929–941. [[CrossRef](#)] [[PubMed](#)]



© 2019 by the author. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (<http://creativecommons.org/licenses/by/4.0/>).