

Chemical and Thermochemical Properties of Graphene Oxide Based Materials: Safety Implications and Applications

By

Yang Qiu

B.S. Peking University, 2007

M.S. Chinese Academy of Sciences, 2010

A DISSERTATION SUBMITTED AS PARTIAL FULFILLMENT OF THE REQUIREMENTS
FOR THE DEGREE OF DOCTOR OF PHILOSOPHY IN ENGINEERING AT
SCHOOL OF ENGINEERING OF BROWN UNIVERSITY

PROVIDENCE, RHODE ISLAND

MAY 2016

© Copyright 2016 by Yang Qiu

This dissertation by Yang Qiu is accepted in its present form by the School of Engineering as satisfying the dissertation requirement for the degree of Doctor of Philosophy.

Date: _____

Professor Robert Hurt, Advisor

Recommended to the Graduate Council

Date: _____

Professor Agnes Kane, Reader

Date: _____

Dr. Indrek Külaots, Reader

Approved by the Graduate Council

Date: _____

Peter Weber, Ph.D. Dean of the Graduate School

Vita

Yang Qiu was born on October 9th, 1984 in Kunming, China. He received his B.S. degree from Peking University in 2007 and M.S. degree from Chinese Academy of Sciences in 2010. After that, he came to Brown University and continued his graduate study in the School of Engineering in 2010. He has been a Superfund Research Program Trainee at Brown SRP Training Core since 2010 and affiliated with the Institute at Brown for Environment and Society. He got Engineer in Training Certification in New Hampshire in 2012. During the doctoral studies at Brown, his research mainly focused on the chemical and thermochemical properties of graphene based materials and their safety implications and applications.

Acknowledgements

First and foremost, I am most grateful to my advisor Prof. Robert H. Hurt. Without his guidance and patient support, I couldn't finish this study. He is a remarkable advisor in research and a helpful mentor in life. I want to thank Prof. Agnes B. Kane. She is incredibly nice and helpful for providing suggestions throughout my research and study. I also want to give special thanks Dr. Indrek Külaots. His expertise in thermal science and sense of humor greatly contribute to the success of this work. He is also one of my best friends at Brown.

Many other people helped me a lot in the last five years and I want to express my sincere gratitude to them all. I would like to thank Dr. Charles Vaslet, Anthony McCormick, Dr. Annette Von Dem Bussche, Zachary Saleeba, Dr. Pranita Katwa, Dr. Hector F. Garces, Dr. Guanglai Li, Benjamin Lyons, Paul Waltz, Joseph Orchard, for their assistance in my research.

My thanks also go to my fellow labmates and good friends. I would like to thank Dr. Yantao Chen, Dr. Jingyu Liu, Dr. Fei Guo, Dr. Megan Creighton, Dr. Xiaoshu Lv, Dr. Po-Yen Chen, Dr. Lorin Jakubek, Zhongying Wang, Ruben Spitz, Jaskiranjeet Sodhi, Alisa Owens, Samuel Moore, and Colin Felton for working with me in teams on different projects and for always being so cheerful.

Last but most importantly, I would like to express my deepest gratitude to my parents for their unconditional love and persistent support throughout my life.

Thank you all!

Abstract

Abstract of CHEMICAL AND THERMOCHEMICAL PROPERTIES OF GRAPHENE OXIDE BASED MATERIALS: SAFETY IMPLICATIONS AND APPLICATIONS by YANG QIU, Ph.D., Brown University, May 2016

The successful application of new materials requires good understanding of their basic properties which govern their behaviors during manufacturing, processing, storage and disposal. Graphene based materials as an emerging family of carbon materials with promising mechanical, electrical and thermal properties have been believed to be “game changer” materials. Among these materials, the most promising category for large-scale production and application in industry are graphene oxide based materials, but some of their properties are still not well understood. Here this thesis focuses on investigating the chemical and thermochemical properties of graphene oxide based materials with both experimental and theoretical approaches. These properties are found closely related to materials chemical structures, more specifically the functional groups of the materials at the molecular level. For example, the conjugated π domain determines their capability to scavenge free radicals; the hydroxyl and carboxyl groups determine the dispersibility in aqueous or polar solvents; the epoxy groups determine the exothermic nature of the graphene oxide pyrolysis; the carbon backbone determines the reducing capability at high temperature. With the knowledge learned in this study, their implications for safe processing and storage were investigated using numeric simulations. New potential applications like oxidation protection using the antioxidant properties of the π conjugated domain and multifunctional platform for theranostics or pollutant cleanup using a couple of the unique properties of graphene oxide have been proposed and explored.

Table of Contents

Vita.....	iv
Acknowledgements.....	v
Abstract.....	vi
Table of Contents	vii
List of Figures.....	xiii
List of Tables	xvi
CHAPTER 1 INTRODUCTION	1
1.1 Graphene oxide.....	2
1.2 The production of GO.....	3
1.3 GO properties and applications	8
1.4 GO based materials.....	9
1.5 GO based materials as important materials for advanced applications	13
1.5.1 GO based materials in composite materials	13
1.5.2 GO based materials for energy storage	15
1.5.3 GO Based materials for sensor application.....	17

1.5.4 GO based materials for biomedical applications	17
1.6 The energetic nature of GO	19
1.7 The scopes of this thesis	19
1.8 References	20
 CHAPTER 2. GRAPHITE OXIDE EXPLOSIVE THERMAL REDUCTION AND SAFETY	
IMPLICATIONS FOR LARGE SCALE STORAGE AND PROCESSING	32
 2.1 Introduction.....	33
2.2 Materials and methods.....	35
2.2.1 Materials	35
2.2.2 Methods.....	36
2.3 Results and Discussion.....	38
2.3.1 Explosive thermal reduction of GO.....	38
2.3.2 Morphology and pore structure of the rGO products.....	42
2.3.3 Factors influencing the thermal reaction mode	45
2.3.3.1 Water	45
2.3.3.2 Impurities.....	48
2.3.3.3 Sample Mass	50
2.3.4 Safety implications	51

2.4 Conclusions	53
2.5 References	54
CHAPTER 3 KINETICS AND THERMOCHEMISTRY OF GRAPHITE OXIDE EXOTHERMIC THERMAL REDUCTION	59
3.1 Introduction.....	60
3.2 Materials and methods.....	62
3.2.1 Materials	62
3.2.2 Characterization	62
3.2.2.1 Morphology.....	62
3.2.2.2 Thermal analysis	63
3.2.3 Decomposition kinetics and thermochemistry analysis	63
3.2.3.1 Decomposition reaction kinetics.....	63
3.2.3.2 Self-heating analysis	64
3.2.3.3 Enthalpy of decomposition analysis	64
3.3 Results and discussion	65
3.3.1 GO thermal exfoliation in nitrogen and air atmospheres	65
3.3.2 Role of salt impurities and reducing agents in GO decomposition reaction	67
3.3.3 Effect of GO aging	72

3.3.4 Results of the decomposition kinetics analysis	74
3.3.5 Discussion of results	75
3.4 Conclusions	79
3.5 References	80
 CHAPTER 4 INTRINSIC CHEMICAL PROPERTIES OF GO-BASED MATERIALS AND THEIR POTENTIAL AS EFFICIENT ANTIOXIDANT	 88
4.1 Introduction.....	89
4.2 Materials and methods.....	91
4.2.1 Materials	91
4.2.2 Characterization	92
4.2.3 Methods.....	92
4.2.3.1 GO based materials preparation.....	92
4.2.3.2 Dye protection assay for •OH scavenging	93
4.2.3.3 •OH scavenging assay by EPR.....	93
4.2.3.4 Non-enzymatic superoxide radical scavenging assay	94
4.2.3.5 Enzymatic superoxide radical scavenging assay.....	94
4.2.3.6 DPPH• and ABTS• ⁺ scavenging assay	94
4.2.3.7 GSH competitive oxidation assay	95

4.2.3.8 MDA assay	96
4.2.3.9 Lipid peroxidation	96
4.2.3.10 TiO ₂ encapsulation by graphene for ROS suppression	97
4.3 Results and discussion	98
4.3.1 Characterization	98
4.3.2 Hydroxyl radical scavenging.....	99
4.3.3 Other radical and oxidant interactions.....	105
4.3.4 Discussion	108
4.4 Conclusions	114
4.5 References	115
CHAPTER 5 GO THERMAL AEROSOL PROCESSING AS A VERSATILE WAY TO BUILD 3D ASSEMBLIES.....	124
5.1 Introduction.....	125
5.2 Materials and methods.....	126
5.2.1 Materials	126
5.2.2 Methods.....	127
5.2.2.1 Synthesis of Au NPs.....	127
5.2.2.2 Synthesis of FeOOH Nanorods:	127

5.2.2.3 Aerosol processing of GO	127
5.2.2.4 Characterization	128
5.2.2.5 MRI/CT/Magnetometry	128
5.3 Results and discussions	131
5.3.1 Thermal aerosol synthesis of crumpled graphene nanosacks	131
5.3.2 Nanosack sealing and controlled release	136
5.3.3 Multimodal imaging	139
5.3.4 Carbothermal reduction as a new route for metal/metal oxides production	145
5.4 Conclusion	148
5.5 References	150
CHAPTER 6 SUMMARY OF THESIS	153

List of Figures

Figure 1.1 GO structural models	3
Figure 1.2 Top down production strategy of GO sheets.	4
Figure 1.3 Purification of GO	6
Figure 1.4 Time-resolved tests for Au-rGO sensor as exposure to multiple air-gas cycles.....	17
Figure 2.1 GO film preparation.....	37
Figure 2.2 Images of DSC cell and TGA sample pan before and after micro-explosions.	39
Figure 2.3 Thermal analysis of the GO reduction process and pore structure of the rGO product.	40
Figure 2.4 Morphology and pore structure of powder produced by explosive thermal reduction of GO cake.	43
Figure 2.5 Effect of interstitial water on the mode of GO thermal reduction (explosive vs. non-explosive)-DSC thermograms	46
Figure 2.6 Role of potassium hydroxide immersion, residual potassium content (K), and total sample mass in the explosive mode of GO reduction.....	47
Figure 2.7 Safety implications of GO explosive reduction.	52
Figure 3.1 DSC thermograms of GO thermal reduction under different condition	66
Figure 3.2 Role of salt impurities on GO decomposition and rGO combustion kinetics.....	68
Figure 3.3 pH effects on the structural and thermal behavior of GO.	70
Figure 3.4 Reduction effects on the thermal behavior of GO.....	72

Figure 3.5 Origin of the exothermicity and aging effects of GO thermal reduction.....	73
Figure 3.6 Quantitative determination of GO thermal decomposition kinetics.....	75
Figure 3.7 Self-heating behavior of large GO samples during storage.....	78
Figure 4.1 GO based materials characterization.....	99
Figure 4.2 GO as a hydroxyl-radical scavenger.....	100
Figure 4.3 GO antioxidant function in systems with $\cdot\text{OH}$ generation by UV-irradiation of hydrogen peroxide.....	102
Figure 4.4 Comparison of different graphene-based materials as $\cdot\text{OH}$ scavengers.....	105
Figure 4.5 GO interactions with other radical species.....	107
Figure 4.6 Graphene material activity against hydrogen peroxide and lipid peroxy radicals.....	108
Figure 4.7 Antioxidant mechanisms and example application.....	112
Figure 5.1 Aerosol-phase synthesis of cargo-filled nanosacks and examples of binary materials with a single homogeneous cargo.....	132
Figure 5.2 Proposed theory for colloidal sack-cargo assembly in binary systems.....	136
Figure 5.3 Information on graphene nanosack sealing and solute release as relevant to delivery functions.....	139
Figure 5.4 Example multifunctional probes as dual CT/MRI contrast agents.....	141
Figure 5.5 Clinical MRI and X-ray CT results showing the contrast performance of G/Fe ₃ O ₄ G/Au and G/Au/Fe ₃ O ₄ multifunctional probes.....	144

Figure 5.6 Carbothermal reduction by the graphene sheets for the synthesis of G/Fe ⁰ nanosacks using G/Fe ₃ O ₄	146
Figure 5.7 Changes of obtained iron-containing rGO nanosacks with increasing temperature for the themal aerosol processing.....	147

List of Tables

Table 2.1 Measured thermochemical parameters of graphene oxide reduction.....	42
Table 2.2 Decomposition enthalpies for graphene oxide and a selection of energetic materials and reactive industrial chemicals known to pose processing safety hazards associated with thermal runaway reactions.....	50

CHAPTER 1 INTRODUCTION

1.1 Graphene oxide

Graphene oxide (GO) is a nonstoichiometric carbon material with various compositions. Though its name sounds like a graphene derived material, it was actually discovered and studied far earlier than the “groundbreaking experiments regarding the two-dimensional material graphene” in 2004.¹⁻² The study on GO can be tracked back to late 1850s when B. Brodie attempted to determine the “atomic weight of graphite”.³

Differed from graphene, the chemical structure of GO is still in controversy though has been studied for decades. In the early pursuit to answer this question, Hofmann and Holst proposed epoxy as the only oxygen functionality and an ideal formula of GO as C_2O .⁴ This model however cannot explain the relatively high hydrogen contents since only edge sites are supposed to bound to hydrogen atoms. Ruess proposed a wrinkled structure with cyclohexane chairs and axial hydroxyl groups and ether oxygens in 1,3 positions.⁵ This model better explained the hydrogen content and also explained the observed acidity by enolic hydroxyl species and carboxylic groups at the edge sites. Later this model was further improved by Scholz and Boehm by removing ether groups and opening some hexane rings since characterizations on GO indicated no presence of ether.⁶ Two decades later Nakajima et al proposed another structure as an analog of poly carbon fluoride, $(C_2F)_n$, based on characterizations on fluorinated GO.⁷ As more advanced experimental techniques have been used in investigating this issue. Researchers gradually recognized that GO does not have determined stoichiometric structure. The most widely accepted model by present researchers is such a model. Randomly distributed oxygen functionalities like epoxyl and hydroxyl cover the six-membered carbon ring sheets, which terminate with carboxyls, ketones and phenolic hydroxyls. This model was proposed by Lerf et al based on a series of

solid-state NMR studies together with XPS and FTIR characterization in late 1990s and early 2000s.⁸⁻⁹

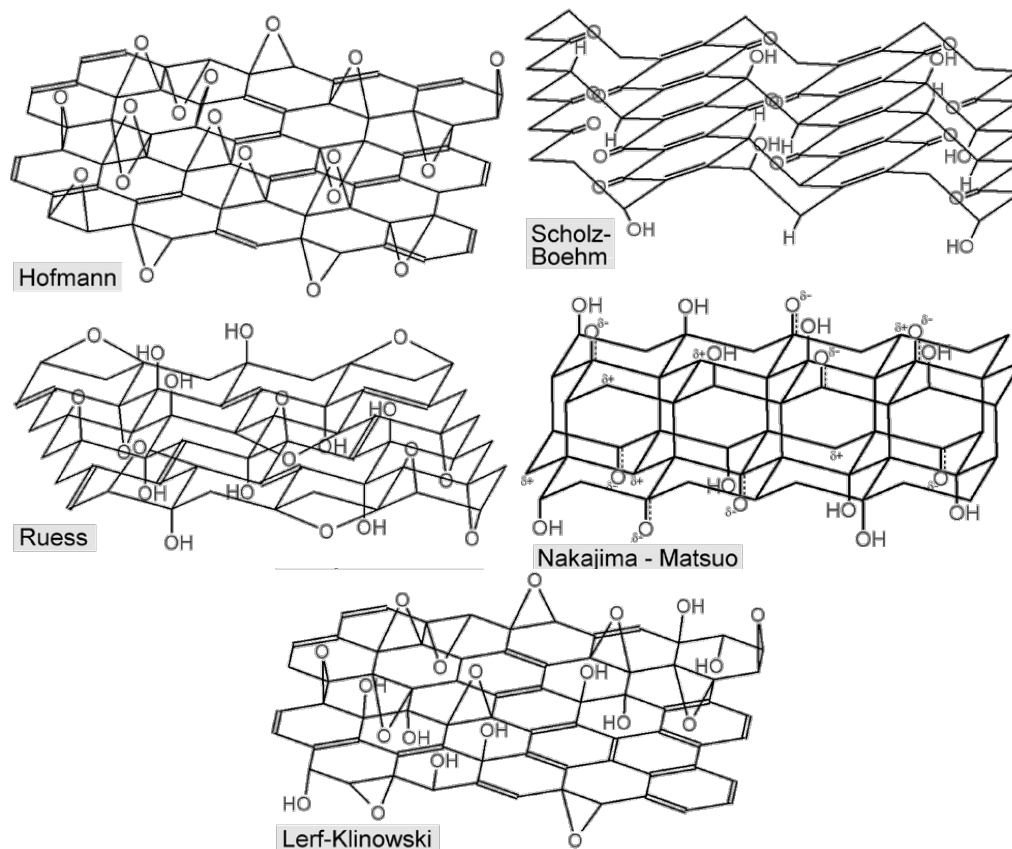


Figure 1.1 GO structural models

Hoffmann et al proposed oxygen species bind to the carbon atoms of the hexagons by epoxy (1,2-ether) linkages with an ideal formula of C_2O . Ruess proposed a wrinkled carbon sheet composed of *trans* linked cyclohexane chairs with the fourth valencies of carbon atoms bound to axial OH groups and ether oxygens in 1,3 positions. Scholtz and Boehm revised the stereochemical structure of Ruess model based on the experimental observations. Nakajima and Matsuo proposed a $(C_2F)_n$ type model by studying the structure of fluorinated GO. Lerf and Klinowski proposed a GO layer as a random distribution of flat aromatic regions with unoxidized benzene rings and wrinkled regions of alicyclic six-membered rings bearing hydroxyl and ether groups with carboxyl and phenolic or aliphatic hydroxyl groups on the edge sites based on NMR studies. This is consistent with most of the experimental observations on GO and is the most accepted model to date. Adapted from *Chemistry of Materials* **2006**¹⁰

1.2 The production of GO

GO can be produced in various ways. Some of them are dramatically different from the others. As other nanomaterials, GO can be produced through either bottom-up or top-down

strategy. Bottom-up strategy is the direct oxidation of monolayer or few layer graphene. It can involve chemical or thermal treatment.¹¹ This strategy has been seldom used either in research or industry. The top-down strategy has been almost exclusively used in the vast majority of GO research and manufacturing. This strategy is essentially exfoliation of oxidized graphite and usually involves three steps as oxidation, purification and exfoliation. The first step is treating graphite with strong oxidizing reagents in liquid or gas phase. The second step is a series of washing, centrifugation and/or filtration to purify and collect the desired product. The third step is achieved mainly through shearing or sonicating the product in polar solvents.

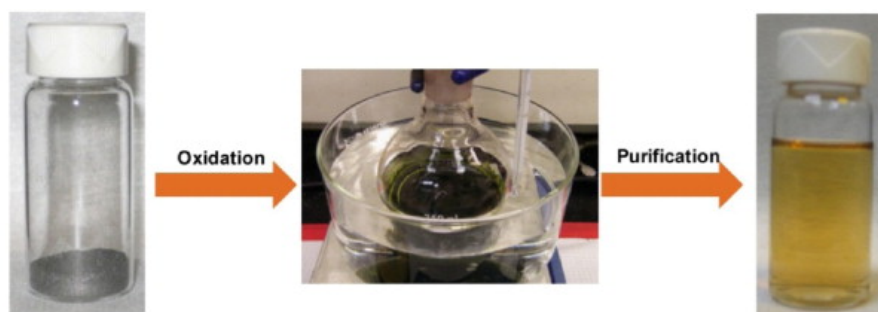


Figure 1.2 Top down production strategy of GO sheets.

Graphite is first oxidized by strong oxidizing agents. This can sometimes involve multistep or repeated oxidation to create satisfactory amount of oxygen functionalities. After extensive purification the final GO product can be dispersed in polar solvents like water. Adapted from *Nano Today* 2012¹²

Solution processing is most widely used to oxidize graphite. There are three most common methods, namely, Brodie method³, Staudenmaier method¹³ and Hummers method¹⁴. In the original research, Brodie attempted to determine the “atomic weight of graphite” and used this oxidizing mixture to treat graphite of Ceylon, the natural occurring vein graphite mined in Sri Lanka. He used a mixture of potassium chlorate (KClO_3) and fuming nitric acid (HNO_3) to treat graphite. Both KClO_3 and fuming HNO_3 are among the strongest oxidizing agents in chemical processing. Fuming HNO_3 is known to be effective to oxidize

graphenic carbon surfaces while KClO_3 is known to generate oxidizer dioxygen.¹⁵ The GO product he obtained has a C:H:O ratio of 61.04 : 1.85 : 37.11.³ The oxidation efficiency of this treatment is not satisfactory. Repeated oxidation treatments are usually necessary to achieve desired oxidation. 40 years later, Staudenmeier improved Brodie method. He proposed adding KClO_3 in multiple aliquots into the oxidation mixture in the presence of concentrated sulfuric acid (H_2SO_4) over the course of reaction. This greatly improves the oxidation efficiency of a single treatment used in Brodie method.¹³ This also allows a one-batch processing to be possible. However, due to safety concerns of the highly toxic and explosive gas byproduct chlorine dioxide (ClO_2), only a small number of researchers produce GO through these two methods. A safer method was developed by Hummers and Offeman in the 1950s. The oxidizing mixture of this method consists of potassium permanganate (KMnO_4), sodium nitrate (NaNO_3) and concentrated H_2SO_4 ¹⁴ and therefore no ClO_2 is produced in this processing. This is a safer and easier method than its two predecessors. Hummers method has since become the most popular one to produce GO in research laboratories.¹⁶ It has been modified and improved a lot in different ways during the last few decades and various versions of modified Hummers methods have been used by different labs. Though modified Hummers methods are more popular in research lab, Staudenmeier method is still being used in industry due to its relatively less tedious purification steps and easier processing procedures.

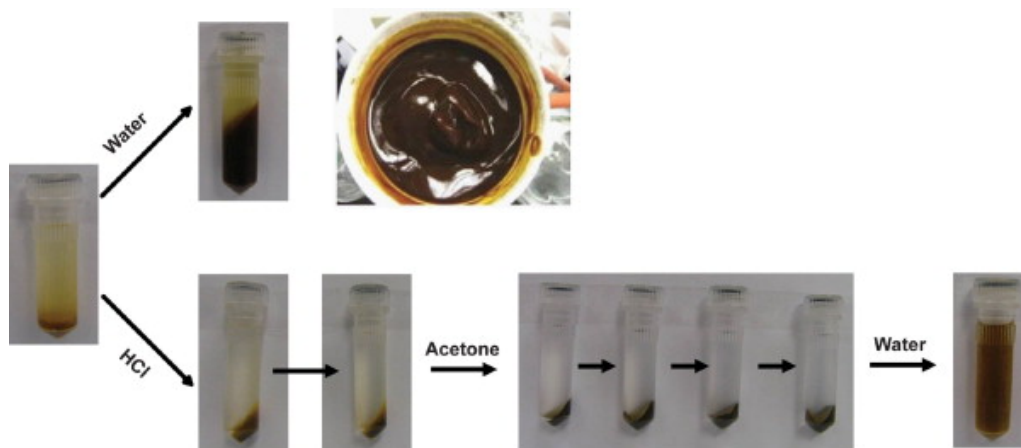


Figure 1.3 Purification of GO

Neither filtration nor centrifugation of GO in water is practical. The flexible sheets can easily stack and quickly block the pores on the membrane during filtration, preventing water going through the membrane. The as prepared GO colloidal suspension is very stable and is thus difficult to pellet with the centrifugal force generated by regular benchtop centrifuge. To overcome this issue, acid (usually HCl) could be added to neutralize the GO surface charge so as to destabilize the colloidal system. This treatment makes centrifugation much easier. The HCl can be washed away by acetone in subsequent purification steps. Adapted from *Nano Today* 2012 ¹²

Purification is usually a challenging step in the process of GO production since GO tends to gel in water, which leads to clogging and blocking membrane during filtration or dialysis.^{3, 12} Ultra high-speed centrifugation is an option but the instrument is costly and is not available for most labs. A more practical way is to make GO colloidal system less stable, which allows GO pelleting with regular benchtop centrifuge. An effective approach is to add acid. GO tends to aggregate at highly acidic environment since the electrical repulsion between individual floating sheets is reduced. A large amount of protons in solution suppresses the deprotonation of hydroxyl and carboxyl groups of GO and in turn shift GO ζ -potential towards neutral. The enhanced ionic strength also contributes to reduce the ζ -potential according to DLVO theory. Hydrochloric acid wash has been proved to be effective to prevent gelation in aqueous phase.¹⁷ Subsequent washing is necessary to remove the anions introduced by acid washing. Acetone is usually used for removing the chloride ions. It however bears threats for the processing safety. As Lee et al reported

residual acetone left on the glassware could be converted by peroxide and sulfuric acid to acetone peroxide, which is a highly explosive substance.¹⁸

GO solids spontaneously disperse in water and form multilayer platelets. To achieve monolayer GO dispersion, exfoliation is usually necessary. It is not a difficult process but still not trivial and can greatly affect the lateral dimension of exfoliated GO sheets. There are numerous ways to exfoliate layered materials, like shear, ultrasonication and thermal expansion. However, GO undergoes rapid chemical change upon heat treatment, thermal expansion is not an option to obtain exfoliated GO. Shearing in solution is a good approach but can be inefficient if the applied force is not strong enough and can potentially take extremely long time to obtain a majority of monolayer. Ultrasonication is an easier and more common choice to obtain monolayer GO in solution but it also has some problems. For example larger sheets can easily be broken into smaller pieces by sonication. This happens even with mild bath sonication.

The composition and properties of GO can vary a lot depending on the oxidative processing. For instance, the C:O ratios of GO produced by Brodie and original Hummers methods are usually higher (indicating less oxidization) than those by Staudenmeier method. Later developed modified Hummers methods greatly improve the oxidation efficiency and could achieve similar or even better oxidation than Staudenmeier method.¹⁶ Moreover, the graphite source and processing conditions also contribute to the great variance of GO properties. The lateral dimension and thickness of the resulting GO largely depend upon the starting graphite and exfoliation process. Smaller graphite flakes and stronger sonication condition are more favorable to produce thinner GO sheets since smaller graphite flakes are more likely to be oxidized to high extent and highly oxidized GO solids are easier to exfoliate. Larger graphite flakes are potentially better choice to produce

GO of larger lateral dimension but larger graphite flakes are at the same time more difficult to oxidize and the obtained GO solids are in turn more difficult to exfoliate. In addition, GO sheets exfoliated by sonication usually have small lateral dimension though it is the most widely used method. To get larger GO sheets, shearing by gentle stirring can be a good choice though the yield of monolayer is low.¹⁹

1.3 GO properties and applications

GO differs from graphene in many ways though both are graphenic two-dimensional materials. Graphene has superior thermal and electrical conductivity while GO does not; graphene is highly hydrophobic while GO is hydrophilic; graphene is an ideal all carbon atom based material while GO contains a large portion of heteroatoms; graphene is chemically inert and stable in atmosphere while GO is reactive with a large range of substances and can even spontaneously change in atmosphere over time; ²⁰ graphene does not have special affinity to water molecules but dry GO is a highly hygroscopic material and automatically adsorbs water in atmosphere.

Due to its unique properties, GO itself have been proposed for a variety of applications. For instance, its hydrophilic oxygen functionalities together with hydrophobic sp² batches on its surface make it an ideal surfactant. It can form stable Pickering emulsions with organic solvents the same way as regular nanoparticles. It can also process hydrophobic materials in water the same way as molecular dispersing agents. ²¹ GO is considered as a two-dimensional random diblock copolymer with a graphitic block and a heavily hydroxylated block. It can spontaneously form liquid crystal structures in aqueous phase under mechanical force ²²⁻²³ and can also guide the assembly of other materials through π - π stacking and hydrogen bonding when incorporated into them.¹²

1.4 GO based materials

GO is also a promising precursor for large-scale manufacturing of graphene-based carbon materials. The traditional and commonly used approaches to produce graphene include micromechanical exfoliation, chemical vapor deposition (CVD) of hydrocarbon, epitaxial growth and colloidal suspension exfoliation. Micromechanical exfoliation, or “Scotch tape” method, is the method Geim et al used in their groundbreaking experiments, which lead to the discovery of graphene. Through repeated peeling of highly oriented pyrolytic graphite (HOPG), a mixture of single layer and few layer graphene can be obtained.^{2, 24} It is apparently difficult to produce a large quantity of high quality graphene through this method. CVD with hydrocarbon is probably the most promising way till now to produce high quality monolayer graphene with low level of defect and large lateral dimension.²⁵ However, it is relatively tedious to transfer the as-prepared graphene thin film from metal substrate onto/into other surface/medium and more importantly the materials yield is not much higher than the “Scotch tape” method and cost is high. Similarly, the graphene epitaxial growth on electrical insulating surfaces such as SiC is achieved through the graphitization of carbide at high temperature under ultra-high vacuum.²⁶⁻²⁷ Although this method produces high quality epitaxial graphene it still suffers similar problems as CVD method. Colloidal suspension exfoliation is relatively new and has been proved to be applicable to a large number of similar two dimensional materials like molybdenum disulfide (MoS_2) and hexagonal boron nitride (h-NB). Organic solvent whose surface energy matches that of graphene can stabilize exposed graphene surfaces generated by agitation like sonication, electric field or shearing, thus allowing accumulation of exfoliated sheets, which is usually unlikely to happen in aqueous solution. Some ionic or nonionic surfactant can achieve similar goal if they can cover the exposed graphene surface and reduce its

surface energy in aqueous phase.²⁸⁻²⁹ This is a very promising method to produce graphene in large scale. The problem for this method is the extremely low yield of single and few layer sheets and the difficulty involved in their separation. The resulting product is usually a mixture of graphene sheets with a great variety of thickness and lateral dimension. Only a small portion of the mixture is one or two layer sheets. The separation is tedious and to achieve better results ultrahigh speed centrifugation and density gradient reagents are usually used.³⁰ To improve the yield, more violent or longer exfoliation can be applied, but the lateral dimension of the graphene produced in this process is very small. Repeated exfoliation of the large/thick sediment after each separation cycle is proved to be helpful to mitigate this issue and to certain extent improve the yield of thin sheets.³⁰ In summary the traditional ways to produce graphene all share similar problem of low yield though they can produce relatively high quality monolayer or few layer graphene. GO reduction nevertheless provides an alternative way to achieve high yield production of graphene-based materials. More strictly speaking, this family of materials should be called GO based materials since their precursors are GO.

Chemical, electrochemical, and physical treatments can be applied to produce GO based materials. These materials can be of various properties and be produced in hundreds of ways, but the majority of them contain reduced GO (rGO). Reduction partially restores the electrical/thermal conductivity and hydrophobicity of pristine graphene for use as a 2D composite filler or conducting film. Chemical reductions are usually easier to tune since reductant concentration determines the reaction rate and the concentration is easy to control, but it could introduce heteroatoms into the final product under many situations. A wide range of reducing agents are known to be applicable to reduce GO. Both strong reductants, such as hydrazine³¹, dimethylhydrazine³², hydroquinone³³ and sodium boron hydride (NaBH₄)³⁴ and weak/mild reductants like glutathione³⁵, ascorbic acid³⁶ and citric

acid³⁷ have been proved to be capable of reducing GO. Electrochemical reduction is usually achieved through depositing GO on electrode which supplies electrons to reduce oxygen functionalities. This approach does not involve the use of reducing agents. It has been demonstrated as an effective way to produce highly conductive rGO products though the details of the mechanism are till not clear. ³⁸ The greatest concern of this method is its inability to scale up to meet industry needs due to the limited amount of GO allowed to be deposited onto the small surface of electrode.

“GO thermal reduction” is the term used in this field to denote GO thermal treatment in inert gas because the main goal is to produce the reduced, graphene-like solid product, rGO. Strictly speaking, this process is not a reduction from the chemical point of view since there is no external reductant (electron donor), but is rather a pyrolysis or chemical disproportionation, in which the original carbon atoms partition into reduced forms in solid rGO and oxidized forms that are primarily carbon oxide gas-phase byproducts (CO, CO₂). Thermal reduction is usually “clean” in terms of the elemental composition of final product and its influence to the environment; however it usually results in restacking which leads to no or little surface area of the final product material.

GO thermal reduction could happen in two modes, mild deoxygenation (non-explosive mode) and violent spontaneous exfoliation (explosive mode). The product of mild deoxygenation normally maintains the morphology of the precursor GO solid. As GO drying leads to massive surface area loss (for example from 2600 to 40 m²g⁻¹) due to alignment and face-to-face restacking³⁹, direct deoxygenation of consolidated GO solids, i.e. GO cake or graphite oxide, will produce rGO cake with similar morphology and small specific surface area. However, if the GO precursor can be dispersed in some solid medium, this method still maintains the initial morphology and keeps the exposed surface of GO sheets during

reduction. This is especially attractive for *in situ* reduction of GO as filler in composite materials as the structure of composite materials should be maintained over the course of the processing. The violent thermal exfoliation is a fast thermal runaway process at elevated temperature when over pressure quickly builds up and overcome the interlayer Van der Waals forces. Depending on the GO initial mass, the sample can generate a large amount of gas products (40 to 50 % of the initial mass) and can produce highly porous products, whose surface area can be much higher than those obtained by mild deoxygenation. Thermal exfoliation of consolidated GO solid is especially attractive for large-scale production of GO-derived few-layer-graphene flakes ⁴⁰, or expanded graphene-based powders with high porosity and surface area for catalysis, separation, or gas storage applications.⁴¹ Thermal exfoliation of multilayer GO for the production of thin graphene flakes has been studied from different aspects in recent years ⁴²⁻⁴⁸. A rapid heating is usually needed to make sure heat and gas generation beat heat loss and gas transport so as to initiate thermal runaway reaction. High vacuum provides extra support for exfoliation by increasing the mechanical driving force for flake expansion, which is the difference between the internal (interstitial) pressure and the environmental pressure. High vacuum can be used for improving low temperature exfoliation efficiency. ^{45, 48} The introduction of hydrogen (H₂) in the surrounding gas phase and pretreatment of GO with hydrochloride (HCl) has also been found to enhance the thermal exfoliation of graphite oxide.⁴⁹ Hydrogen in the gas environment during reduction can violently react with hydroxyls on GO, and induce thermal decomposition.⁴⁹ HCl is a volatile species. Under thermal treatment, the rapid gasification and release of intercalated HCl molecules create additional overpressure needed to successfully overcome interlayer Van der Waals forces. Thermal exfoliation in GO induces the decomposition of epoxides and hydroxyls, the reaction rate of which competes with the diffusion of their gas products CO₂ and CO. Successful thermal exfoliation is

achieved when decomposition rate of GO overwhelms the mass transfer of gas products so that the internal pressure can build up to reach the threshold overpressure.⁵⁰ Increasing O/C ratio of GO⁴³ is beneficial for higher GO decomposition rate and therefore, enhances thermal exfoliation due to the higher density of reactive sites. For the successful thermal exfoliation overpressure is needed to overcome the van der Waals forces in the adjacent GO layers.⁵⁰

1.5 GO based materials as important materials for advanced applications

Due to the large amount of defects introduced during the oxidation, GO based materials are in general not ideal materials for building electronic devices like field effect transistors, but their good stability to maintain exfoliated state and the ease to scale up their production make them ideal starting materials to produce various types of graphene-based materials for other applications. Most GO based materials in application are in reduced form. Therefore the following discussion will focus on this type of materials while non-reduced form will also be mentioned.

1.5.1 GO based materials in composite materials

Thanks to its good electrical conductivity, reduced GO based materials are considered good candidate to be incorporated into the matrix of other materials to produce composite. As compared with the solution process exfoliated graphite, the aspect ratio (lateral dimension: thickness) of individual rGO sheet can be high if processed properly and the size distribution can be better controlled, which is crucial for achieving lower percolation threshold when incorporated into composite⁵¹⁻⁵³. Processing condition largely determines

the product's electrical and mechanical properties. If reduction precedes the introduction of the stabilizing matrix like polymer, severe aggregation will happen and compromise the performance of the composite. An alternative and better strategy is to incorporate GO into polymer before reduction. As reported by Stankovich et al, thoroughly mixing exfoliated GO with polystyrene and phenyl isocyanate in dimethylformamide(DMF) and subsequently adding reducing agent dimethylhydrazine at 80 °C for 24 h can produce a composite with rGO single sheets uniformly distributed in the polystyrene matrix without obvious stacking.³² In addition to being used as filler in a composite, rGO can also be used as a support to anchor semiconductor or metal nanoparticles to achieve various types of functions, in particular, sensor and catalysis. Either *in situ* synthesizing nanoparticles when GO was reduced or attaching pre-synthesized nanoparticles onto functionalized or unfunctionalized rGO sheets can produce this type of composites. Muszynski et al developed a way to synthesize gold nanoparticles at the same time when reducing GO using NaBH₄.⁵⁴ The resulting composite maintains the surface plasma resonance feature of gold nanoparticles. Yang et al reported synthesis of superparamagnetic Fe₃O₄-graphene oxide composite through *in situ* co-precipitation⁵⁵. As an alternative strategy, He et al functionalized Fe₃O₄ nanoparticles with tetraethyl orthosilicate and (3-aminopropyl) triethoxysilane and then covalently bind the nanoparticles onto the 1-ethyl-3-(3-dimethylaminopropyl) carbodiimide and N-hydroxysuccinnimide activated GO surface⁵⁶. Williams et al used UV light to reduce GO in the presence of TiO₂ nanoparticles to produce TiO₂-graphene composite. In this system the as-synthesized TiO₂ nanoparticles spontaneously bind to the carboxyl groups on GO surface and donate electrons to reduce GO upon UV irradiation.⁵⁷

1.5.2 GO based materials for energy storage

A great variety of GO based materials have been synthesized for energy storage. One important application is in lithium battery. GO based materials are good anode materials due to their large specific surface area and their capability to facilitate electron and hole transfer on the surface. The traditional rechargeable lithium ion battery electrode material graphite has a theoretical maximum lithium insertion capacity of 372 mAh/g by forming LiC_6 .⁵⁸ However, the theoretical insertion capacity for monolayer graphene as electrode material is 744 mAh/g when forming Li_2C_6 .⁵⁹⁻⁶⁰ The experimentally measured capacity of graphene nanosheet electrode is 540 mAh/g which is much higher than the graphite electrode. When carbon nanotube or fullerene is incorporated into the electrode material, the capacity can be further improved to 730 or 784 mAh/g, respectively.⁶¹ Functional materials can further improve GO based materials' battery capacity. The battery using silicon-rGO composite as electrode materials shows capacity of more than 2200 mAh/g as reported by Lee, J. et al.⁶² Another important energy storage application is super capacitor. GO based materials may be used to build electrical double layer capacitors and pseudo-capacitors. Capacitance of electrical double layer capacitors is generated by electrostatic charge accumulation at the interface of electrode and electrolyte. Porous carbon materials have long been used for this type of capacitors. The most widely used carbon material is activated carbon or template carbon; however, though these types of materials have high specific surface areas they suffer from problems like uncontrollable random porous structures, which reduce the capacitance. The pore structures of rGO based electrical double layer capacitors are easier to control and the intrinsic high electron mobility of graphene sheets is also beneficial for obtaining high capacitance.⁶³ One of the most promising super capacitors was developed with high vacuum assisted low

temperature (~ 200 °C) exfoliated rGO with a reported capacitance of 264 F/g.⁴⁵ The relatively low temperature used here as compared with the common used thermal exfoliation temperature (above 1000 °C) minimizes new defects introduced in the processing and maintains the integrity of individual precursor sheets, which is important for achieving high capacitance.⁴⁵ The advantage of this processing is its great potential for the scale up of production. Another promising super capacitor was developed by Fan, Z et al. The capacitance of this carefully designed and constructed CNT-graphene sandwich structure was measured to be 385 F/g, which is among the highest capacitances reported to date.⁶⁴ Pseudo-capacitor does not generate electrical capacitance; however, the energy is stored via faradic processes like redox and adsorption. The contribution of adsorption pseudo capacitance is much smaller than redox pseudo capacitance and therefore pseudo capacitance is often called redox capacitance. Carbon material itself is not a pseudo capacitor material since it does not have reversible redox properties under normal conditions of electrochemical process. Pseudo capacitor materials are usually conducting polymers and transition metal oxides or hydroxide which have high theoretical capacitance and intrinsically fast and reversible redox reactions, but the pure functioning materials suffers from low conductivity at neutral condition and low specific surface area, which lowers the effective capacitance and causes poor cycling stability.⁶⁵ GO based materials can improve their performance by providing good conductive support and maintaining high specific surface area for these pseudo capacitor materials. One of the highest capacitance achieved to date was in the study by Yan, J. et al. The rGO-polyaniline composite as prepared by *in situ* polymerization of aniline on rGO nanosheets has 1046 F/g capacitance as compared with 115 F/g capacitance of the pure polyaniline.⁶⁶ GO based materials have also been used in many other energy storage applications like electrocatalysis in fuel cells,⁶⁷ photocatalysis for hydrogen production,⁶⁸⁻⁶⁹ and solar cells.⁷⁰

1.5.3 GO Based materials for sensor application

GO based materials are also promising materials for sensor applications. For example, rGO itself can act as gas sensor. It responds to H_2 and CO gas adsorption by changing conductivity.⁷¹ When coupled with gold nanoparticles, rGO can also detect gas molecules by changing gold nanoparticles color as an optical sensor.⁷²

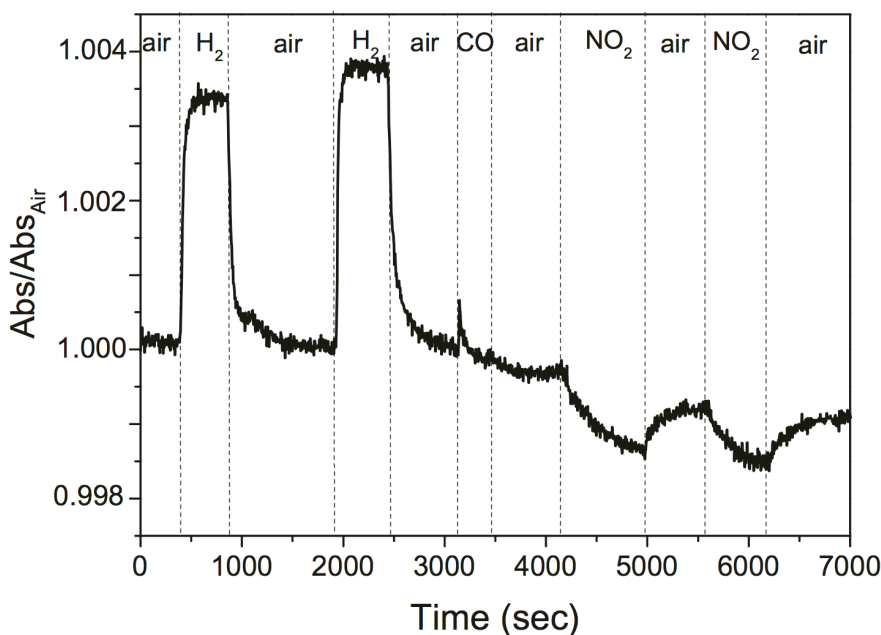


Figure 1.4 Time-resolved tests for Au-rGO sensor as exposure to multiple air-gas cycles

The Au-rGO sensor responds to reducing gas H_2 and CO by increasing optical absorption and to oxidizing gas NO_2 by reducing optical absorption. Adapted from *Carbon* 2014 ⁷²

1.5.4 GO based materials for biomedical applications

GO based materials also have wide applications in biomedical fields. In addition to biosensors, they have also been used for molecular imaging, tissue engineering and drug/gene delivery. In the early efforts, graphene materials were conjugated with fluorescent molecules or quantum dots and were introduced into cells for intracellular

imaging.⁷³⁻⁷⁴ Fluorescence imaging however was later found challenging since graphene materials quench fluorescence. Other options include radioactive and magnetic labeling which enables positron emission tomography (PET) or magnetic resonance imaging (MRI).⁷⁵⁻⁷⁶ Stem cell differentiation plays a key role in building new tissues or even organs in the field of tissue engineering. Chen, G. et al reported GO can support the growth of induced pluripotent stem cells (iPSC) as a substrate and the attachment and differentiation of iPSC can be tuned by changing the reduction state of GO.⁷⁷ Drug delivery has also been demonstrated with functionalized rGO and other GO based materials. Numerous chemotherapy drugs like doxorubicin⁷⁸ and SN38⁷⁹ have been successfully conjugated to the carrier graphene sheets to achieve higher cytotoxicity towards specific targets like cancer cells. It is also possible to simultaneously conjugate multiple drugs onto GO based materials to achieve synergistic therapeutic effects.⁸⁰ In addition to regular chemotherapy drugs, more sophisticated therapeutic functions have also been coupled with rGO carrier. For instance, photosensitizer can be introduced to enable photodynamic therapy.⁸¹ Also, the intrinsic near infrared photothermal effect of graphene sheets makes photothermal therapy possible even without photothermal sensitizer. When combined with chemotherapy, GO based materials can be used for multi-mode synergetic therapy.⁸² Moreover, controlled release as an important function for drug delivery has also been demonstrated when sensitive polymers are grafted onto the carrier. pH- or thermal-sensitive release systems have been developed with Pluronic F127 and poly(N-isopropylacrylamide), respectively.⁸³⁻⁸⁴ There are also growing interests on graphene based gene delivery in recent years. For instance, plasmid DNA delivery has been demonstrated through electrostatically binding DNA on the PEI-GO nanosheets.⁸⁵

1.6 The energetic nature of GO

As a facile route to produce rGO, thermal reduction however involves safety concerns. GO has long been known as thermally unstable, energetic material^{3, 12, 50, 86-94}. 50 years ago Boehm et al. reported that when “heated quickly to about 180 C it deflagrates with production of high volume per mass fluffy carbon” consisting of relatively “thin carbon sheet platelets”.⁸⁸ Nanoscale ribbons of GO made from the oxidative unzipping of carbon nanotubes can also undergo explosive decomposition if heated in N₂ gas.⁹³ Kim et al.⁹⁰ and Krishnan et al.¹² reported the spontaneous ignition of rGO films in air under the influence of potassium hydroxide that act as a catalyst for the carbon combustion. These energetic properties are largely due to the exothermic nature of GO thermal reduction. It is relatively unusual among the oxygen rich organic materials (GO can be seen as an organic material due to its large portion of oxygen functionalities).⁹⁵⁻⁹⁹ GO thermal reduction is typically conducted in the absence of air, either in vacuum or inert gas to protect the rGO product. Under these inert conditions there have been observations of the exfoliation behavior^{40,50}, but no systematic studies on the factors that determine whether a given sample will exhibit the explosive or non-explosive mode.

1.7 The scopes of this thesis

Here in this thesis the energetic thermal reduction of GO will be studied in a more systematic approach so as to develop a theory to address these issues. We hypothesized that the energetic nature of graphene oxide is due to its special chemical structure and must be understood and characterized for the safe handling of GO, especially at large scale. In the following chapters, key factors that could determine the reaction mode (explosive or

non-explosive) of thermal reduction are investigated. To further understand the mechanism of the exothermic thermal reduction process the underlying kinetics and thermochemistry of the non-explosive mode thermal reduction are also studied. In addition, the influence of thermal treatment on the chemical behavior of GO based materials in biological system will be studied with special focus on their oxidative and antioxidative activities. Lastly, the application of thermal treatment on different types of GO based materials are also studied and demonstrated to be a useful approach to better control the property and function of GO based materials or even used as a facile way to produce sophisticated or challenging nano- or microscale systems.

1.8 References

1. The Nobel Prize in Physics 2010. http://www.nobelprize.org/nobel_prizes/physics/laureates/2010/press.html (accessed 7/1/2015).
2. Novoselov, K. S.; Geim, A. K.; Morozov, S. V.; Jiang, D.; Zhang, Y.; Dubonos, S. V.; Grigorieva, I. V.; Firsov, A. A., Electric field effect in atomically thin carbon films. *Science* 2004, 306 (5696), 666-669.
3. Brodie, B. C., On the atomic weight of graphite. *Philos. Trans. R. Soc. London* 1859, 249-259.
4. Hofmann, U.; Holst, R., Über die Säurenatur und die Methylierung von Graphitoxyd. *Berichte der deutschen chemischen Gesellschaft (A and B Series)* 1939, 72 (4), 754-771.

5. Ruess, G., Über das Graphitoxhydroxyd (Graphitoxyd). *Monatshefte für Chemie und verwandte Teile anderer Wissenschaften* 1947, 76 (3-5), 381-417.
6. Scholz, W.; Boehm, H. P., Untersuchungen am Graphitoxid. VI. Betrachtungen zur Struktur des Graphitoxids. *Z. Anorg. Allg. Chem.* 1969, 369 (3-6), 327-340.
7. Nakajima, T.; Mabuchi, A.; Hagiwara, R., A New Structure Model of Graphite Oxide. *Carbon* 1988, 26 (3), 357-361.
8. He, H. Y.; Riedl, T.; Lerf, A.; Klinowski, J., Solid-state NMR studies of the structure of graphite oxide. *J. Phys. Chem.* 1996, 100 (51), 19954-19958.
9. Lerf, A.; He, H. Y.; Forster, M.; Klinowski, J., Structure of graphite oxide revisited. *J. Phys. Chem. B* 1998, 102 (23), 4477-4482.
10. Szabo, T.; Berkesi, O.; Forgo, P.; Josepovits, K.; Sanakis, Y.; Petridis, D.; Dekany, I., Evolution of surface functional groups in a series of progressively oxidized graphite oxides. *Chem. Mater.* 2006, 18 (11), 2740-2749.
11. Liu, L.; Ryu, S. M.; Tomasik, M. R.; Stolyarova, E.; Jung, N.; Hybertsen, M. S.; Steigerwald, M. L.; Brus, L. E.; Flynn, G. W., Graphene oxidation: Thickness-dependent etching and strong chemical doping. *Nano Lett.* 2008, 8 (7), 1965-1970.
12. Krishnan, D.; Kim, F.; Luo, J. Y.; Cruz-Silva, R.; Cote, L. J.; Jang, H. D.; Huang, J. X., Energetic graphene oxide: Challenges and opportunities. *Nano Today* 2012, 7 (2), 137-152.
13. Staudenmaier, L., Verfahren zur darstellung der graphitsäure. *Berichte der deutschen chemischen Gesellschaft* 1898, 31 (2), 1481-1487.
14. Hummers, W. S.; Offeman, R. E., Preparation of Graphitic Oxide. *J. Am. Chem. Soc.* 1958, 80 (6), 1339-1339.
15. Dreyer, D. R.; Park, S.; Bielawski, C. W.; Ruoff, R. S., The chemistry of graphene oxide. *Chem. Soc. Rev.* 2010, 39 (1), 228-240.

16. Compton, O. C.; Nguyen, S. T., Graphene Oxide, Highly Reduced Graphene Oxide, and Graphene: Versatile Building Blocks for Carbon-Based Materials. *Small* 2010, 6 (6), 711-723.
17. He, H. Y.; Klinowski, J.; Forster, M.; Lerf, A., A new structural model for graphite oxide. *Chem. Phys. Lett.* 1998, 287 (1-2), 53-56.
18. Lee, S.; Oh, J.; Ruoff, R. S.; Park, S., Residual acetone produces explosives during the production of graphite oxide. *Carbon* 2012, 50 (3), 1442-1444.
19. Jung, I.; Field, D. A.; Clark, N. J.; Zhu, Y. W.; Yang, D. X.; Piner, R. D.; Stankovich, S.; Dikin, D. A.; Geisler, H.; Ventrice, C. A.; Ruoff, R. S., Reduction Kinetics of Graphene Oxide Determined by Electrical Transport Measurements and Temperature Programmed Desorption. *J Phys Chem C* 2009, 113 (43), 18480-18486.
20. Kim, S.; Zhou, S.; Hu, Y. K.; Acik, M.; Chabal, Y. J.; Berger, C.; de Heer, W.; Bongiorno, A.; Riedo, E., Room-temperature metastability of multilayer graphene oxide films. *Nat Mater* 2012, 11 (6), 544-549.
21. Kim, J.; Cote, L. J.; Kim, F.; Yuan, W.; Shull, K. R.; Huang, J. X., Graphene Oxide Sheets at Interfaces. *J. Am. Chem. Soc.* 2010, 132 (23), 8180-8186.
22. Kim, J. E.; Han, T. H.; Lee, S. H.; Kim, J. Y.; Ahn, C. W.; Yun, J. M.; Kim, S. O., Graphene Oxide Liquid Crystals. *Angew Chem Int Edit* 2011, 50 (13), 3043-3047.
23. Xu, Z.; Gao, C., Aqueous Liquid Crystals of Graphene Oxide. *ACS nano* 2011, 5 (4), 2908-2915.
24. Novoselov, K. S.; Jiang, D.; Schedin, F.; Booth, T. J.; Khotkevich, V. V.; Morozov, S. V.; Geim, A. K., Two-dimensional atomic crystals. *Proc. Natl. Acad. Sci. U. S. A.* 2005, 102 (30), 10451-10453.
25. Eizenberg, M.; Blakely, J. M., Carbon Monolayer Phase Condensation on Ni(111). *Surf. Sci.* 1979, 82 (1), 228-236.

26. Aizawa, T.; Souda, R.; Otani, S.; Ishizawa, Y.; Oshima, C., Anomalous Bond of Monolayer Graphite on Transition-Metal Carbide Surfaces. *Phys. Rev. Lett.* 1990, **64** (7), 768-771.
27. Berger, C.; Song, Z. M.; Li, X. B.; Wu, X. S.; Brown, N.; Naud, C.; Mayou, D.; Li, T. B.; Hass, J.; Marchenkov, A. N.; Conrad, E. H.; First, P. N.; de Heer, W. A., Electronic confinement and coherence in patterned epitaxial graphene. *Science* 2006, **312** (5777), 1191-1196.
28. Nicolosi, V.; Chhowalla, M.; Kanatzidis, M. G.; Strano, M. S.; Coleman, J. N., Liquid Exfoliation of Layered Materials. *Science* 2013, **340** (6139), 1420-+.
29. Smith, R. J.; King, P. J.; Lotya, M.; Wirtz, C.; Khan, U.; De, S.; O'Neill, A.; Duesberg, G. S.; Grunlan, J. C.; Moriarty, G.; Chen, J.; Wang, J. Z.; Minett, A. I.; Nicolosi, V.; Coleman, J. N., Large-Scale Exfoliation of Inorganic Layered Compounds in Aqueous Surfactant Solutions. *Adv. Mater.* 2011, **23** (34), 3944-+.
30. Green, A. A.; Hersam, M. C., Solution Phase Production of Graphene with Controlled Thickness via Density Differentiation. *Nano Lett.* 2009, **9** (12), 4031-4036.
31. Stankovich, S.; Dikin, D. A.; Piner, R. D.; Kohlhaas, K. A.; Kleinhammes, A.; Jia, Y.; Wu, Y.; Nguyen, S. T.; Ruoff, R. S., Synthesis of graphene-based nanosheets via chemical reduction of exfoliated graphite oxide. *Carbon* 2007, **45** (7), 1558-1565.
32. Stankovich, S.; Dikin, D. A.; Dommett, G. H. B.; Kohlhaas, K. M.; Zimney, E. J.; Stach, E. A.; Piner, R. D.; Nguyen, S. T.; Ruoff, R. S., Graphene-based composite materials. *Nature* 2006, **442** (7100), 282-286.
33. Wang, G. X.; Yang, J.; Park, J.; Gou, X. L.; Wang, B.; Liu, H.; Yao, J., Facile synthesis and characterization of graphene nanosheets. *J Phys Chem C* 2008, **112** (22), 8192-8195.
34. Si, Y.; Samulski, E. T., Synthesis of water soluble graphene. *Nano Lett.* 2008, **8** (6), 1679-1682.

35. Pham, T. A.; Kim, J. S.; Kim, J. S.; Jeong, Y. T., One-step reduction of graphene oxide with L-glutathione. *Colloids and Surfaces a-Physicochemical and Engineering Aspects* 2011, *384* (1-3), 543-548.
36. Zhang, J.; Yang, H.; Shen, G.; Cheng, P.; Zhang, J.; Guo, S., Reduction of graphene oxide vial-ascorbic acid. *Chem. Commun.* 2010, *46* (7), 1112-1114.
37. Zhang, Z.; Chen, H. H.; Xing, C. Y.; Guo, M. Y.; Xu, F. G.; Wang, X. D.; Gruber, H.; Zhang, B. L.; Tang, J. L., Sodium Citrate: A Universal Reducing Agent for Reduction/Decoration of Graphene Oxide with Au Nanoparticles. *Nano Res* 2011, *4* (6), 599-611.
38. Zhou, M.; Wang, Y. L.; Zhai, Y. M.; Zhai, J. F.; Ren, W.; Wang, F. A.; Dong, S. J., Controlled Synthesis of Large-Area and Patterned Electrochemically Reduced Graphene Oxide Films. *Chem-Eur J* 2009, *15* (25), 6116-6120.
39. Guo, F.; Creighton, M.; Chen, Y.; Hurt, R.; Külaots, I., Porous structures in stacked, crumpled and pillared graphene-based 3D materials. *Carbon* 2014, *66* (0), 476-484.
40. Zhang, C.; Lv, W.; Xie, X.; Tang, D.; Liu, C.; Yang, Q.-H., Towards low temperature thermal exfoliation of graphite oxide for graphene production. *Carbon* 2013, *62*, 11-24.
41. Park, S.; Ruoff, R. S., Chemical methods for the production of graphenes. *Nat Nanotechnol* 2009, *4* (4), 217-224.
42. Du, Q. L.; Zheng, M. B.; Zhang, L. F.; Wang, Y. W.; Chen, J. H.; Xue, L. P.; Dai, W. J.; Ji, G. B.; Cao, J. M., Preparation of functionalized graphene sheets by a low-temperature thermal exfoliation approach and their electrochemical supercapacitive behaviors. *Electrochim. Acta* 2010, *55* (12), 3897-3903.
43. Jin, M.; Jeong, H. K.; Kim, T. H.; So, K. P.; Cui, Y.; Yu, W. J.; Ra, E. J.; Lee, Y. H., Synthesis and systematic characterization of functionalized graphene sheets generated by thermal exfoliation at low temperature. *Journal of Physics D-Applied Physics* 2010, *43* (27).

44. Jung, I.; Dikin, D. A.; Piner, R. D.; Ruoff, R. S., Tunable electrical conductivity of individual graphene oxide sheets reduced at "low" temperatures. *Nano Lett.* 2008, 8 (12), 4283-4287.
45. Lv, W.; Tang, D. M.; He, Y. B.; You, C. H.; Shi, Z. Q.; Chen, X. C.; Chen, C. M.; Hou, P. X.; Liu, C.; Yang, Q. H., Low-Temperature Exfoliated Graphenes: Vacuum-Promoted Exfoliation and Electrochemical Energy Storage. *ACS nano* 2009, 3 (11), 3730-3736.
46. Schniepp, H. C.; Li, J. L.; McAllister, M. J.; Sai, H.; Herrera-Alonso, M.; Adamson, D. H.; Prud'homme, R. K.; Car, R.; Saville, D. A.; Aksay, I. A., Functionalized single graphene sheets derived from splitting graphite oxide. *J. Phys. Chem. B* 2006, 110 (17), 8535-8539.
47. Ye, J.; Zhang, H. Y.; Chen, Y. M.; Cheng, Z. D.; Hu, L.; Ran, Q. Y., Supercapacitors based on low-temperature partially exfoliated and reduced graphite oxide. *J. Power Sources* 2012, 212, 105-110.
48. Zhang, H. B.; Wang, J. W.; Yan, Q.; Zheng, W. G.; Chen, C.; Yu, Z. Z., Vacuum-assisted synthesis of graphene from thermal exfoliation and reduction of graphite oxide. *J. Mater. Chem.* 2011, 21 (14), 5392-5397.
49. Shen, B.; Lu, D. D.; Zhai, W. T.; Zheng, W. G., Synthesis of graphene by low-temperature exfoliation and reduction of graphite oxide under ambient atmosphere. *Journal of Materials Chemistry C* 2013, 1 (1), 50-53.
50. McAllister, M. J.; Li, J. L.; Adamson, D. H.; Schniepp, H. C.; Abdala, A. A.; Liu, J.; Herrera-Alonso, M.; Milius, D. L.; Car, R.; Prud'homme, R. K.; Aksay, I. A., Single sheet functionalized graphene by oxidation and thermal expansion of graphite. *Chem. Mater.* 2007, 19 (18), 4396-4404.

51. Jing, X.; Zhao, W.; Lan, L., The effect of particle size on electric conducting percolation threshold in polymer/conducting particle composites. *J. Mater. Sci. Lett.* 2000, *19* (5), 377-379.
52. Gokturk, H. S.; Fiske, T. J.; Kalyon, D. M., Effects of Particle-Shape and Size Distributions on the Electrical and Magnetic-Properties of Nickel/Polyethylene Composites. *J. Appl. Polym. Sci.* 1993, *50* (11), 1891-1901.
53. Chodak, I.; Krupa, I., "Percolation effect" and mechanical behavior of carbon black filled polyethylene. *J. Mater. Sci. Lett.* 1999, *18* (18), 1457-1459.
54. Muszynski, R.; Seger, B.; Kamat, P. V., Decorating graphene sheets with gold nanoparticles. *J Phys Chem C* 2008, *112* (14), 5263-5266.
55. Yang, X. Y.; Zhang, X. Y.; Ma, Y. F.; Huang, Y.; Wang, Y. S.; Chen, Y. S., Superparamagnetic graphene oxide-Fe₃O₄ nanoparticles hybrid for controlled targeted drug carriers. *J. Mater. Chem.* 2009, *19* (18), 2710-2714.
56. He, F. A.; Fan, J. T.; Ma, D.; Zhang, L. M.; Leung, C.; Chan, H. L., The attachment of Fe₃O₄ nanoparticles to graphene oxide by covalent bonding. *Carbon* 2010, *48* (11), 3139-3144.
57. Williams, G.; Seger, B.; Kamat, P. V., TiO₂-graphene nanocomposites. UV-assisted photocatalytic reduction of graphene oxide. *ACS nano* 2008, *2* (7), 1487-1491.
58. Winter, M.; Besenhard, J. O.; Spahr, M. E.; Novak, P., Insertion electrode materials for rechargeable lithium batteries. *Adv. Mater.* 1998, *10* (10), 725-763.
59. Dahn, J. R.; Zheng, T.; Liu, Y. H.; Xue, J. S., Mechanisms for Lithium Insertion in Carbonaceous Materials. *Science* 1995, *270* (5236), 590-593.
60. Liu, Y. H.; Xue, J. S.; Zheng, T.; Dahn, J. R., Mechanism of lithium insertion in hard carbons prepared by pyrolysis of epoxy resins. *Carbon* 1996, *34* (2), 193-200.

61. Yoo, E.; Kim, J.; Hosono, E.; Zhou, H.; Kudo, T.; Honma, I., Large reversible Li storage of graphene nanosheet families for use in rechargeable lithium ion batteries. *Nano Lett.* 2008, 8 (8), 2277-2282.
62. Lee, J. K.; Smith, K. B.; Hayner, C. M.; Kung, H. H., Silicon nanoparticles-graphene paper composites for Li ion battery anodes. *Chem. Commun.* 2010, 46 (12), 2025-2027.
63. Sun, Y.; Wu, Q.; Shi, G., Graphene based new energy materials. *Energy Environ. Sci.* 2011, 4 (4), 1113-1132.
64. Fan, Z. J.; Yan, J.; Zhi, L. J.; Zhang, Q.; Wei, T.; Feng, J.; Zhang, M. L.; Qian, W. Z.; Wei, F., A Three-Dimensional Carbon Nanotube/Graphene Sandwich and Its Application as Electrode in Supercapacitors. *Adv. Mater.* 2010, 22 (33), 3723-+.
65. Hall, P. J.; Mirzaeian, M.; Fletcher, S. I.; Sillars, F. B.; Rennie, A. J. R.; Shitta-Bey, G. O.; Wilson, G.; Cruden, A.; Carter, R., Energy storage in electrochemical capacitors: designing functional materials to improve performance. *Energy Environ. Sci.* 2010, 3 (9), 1238-1251.
66. Yan, J.; Wei, T.; Shao, B.; Fan, Z. J.; Qian, W. Z.; Zhang, M. L.; Wei, F., Preparation of a graphene nanosheet/polyaniline composite with high specific capacitance. *Carbon* 2010, 48 (2), 487-493.
67. Kou, R.; Shao, Y. Y.; Wang, D. H.; Engelhard, M. H.; Kwak, J. H.; Wang, J.; Viswanathan, V. V.; Wang, C. M.; Lin, Y. H.; Wang, Y.; Aksay, I. A.; Liu, J., Enhanced activity and stability of Pt catalysts on functionalized graphene sheets for electrocatalytic oxygen reduction. *Electrochem. Commun.* 2009, 11 (5), 954-957.
68. Lightcap, I. V.; Kosel, T. H.; Kamat, P. V., Anchoring Semiconductor and Metal Nanoparticles on a Two-Dimensional Catalyst Mat. Storing and Shuttling Electrons with Reduced Graphene Oxide. *Nano Lett.* 2010, 10 (2), 577-583.

69. Yeh, T. F.; Syu, J. M.; Cheng, C.; Chang, T. H.; Teng, H. S., Graphite Oxide as a Photocatalyst for Hydrogen Production from Water. *Adv. Funct. Mater.* 2010, 20 (14), 2255-2262.
70. Wang, X.; Zhi, L. J.; Mullen, K., Transparent, conductive graphene electrodes for dye-sensitized solar cells. *Nano Lett.* 2008, 8 (1), 323-327.
71. Arsat, R.; Breedon, M.; Shafiei, M.; Spizziri, P. G.; Gilje, S.; Kaner, R. B.; Kalantar-Zadeh, K.; Wlodarski, W., Graphene-like nano-sheets for surface acoustic wave gas sensor applications. *Chem. Phys. Lett.* 2009, 467 (4-6), 344-347.
72. Cittadini, M.; Bersani, M.; Perrozzi, F.; Ottaviano, L.; Wlodarski, W.; Martucci, A., Graphene oxide coupled with gold nanoparticles for localized surface plasmon resonance based gas sensor. *Carbon* 2014, 69, 452-459.
73. Peng, C.; Hu, W. B.; Zhou, Y. T.; Fan, C. H.; Huang, Q., Intracellular Imaging with a Graphene-Based Fluorescent Probe. *Small* 2010, 6 (15), 1686-1692.
74. Chen, M. L.; Liu, J. W.; Hu, B.; Chen, M. L.; Wang, J. H., Conjugation of quantum dots with graphene for fluorescence imaging of live cells. *Analyst* 2011, 136 (20), 4277-4283.
75. Hong, H.; Yang, K.; Zhang, Y.; Engle, J. W.; Feng, L. Z.; Yang, Y. A.; Nayak, T. R.; Goel, S.; Bean, J.; Theuer, C. P.; Barnhart, T. E.; Liu, Z.; Cai, W. B., In Vivo Targeting and Imaging of Tumor Vasculature with Radiolabeled, Antibody-Conjugated Nanographene. *ACS nano* 2012, 6 (3), 2361-2370.
76. Yang, K.; Hu, L. L.; Ma, X. X.; Ye, S. Q.; Cheng, L.; Shi, X. Z.; Li, C. H.; Li, Y. G.; Liu, Z., Multimodal Imaging Guided Photothermal Therapy using Functionalized Graphene Nanosheets Anchored with Magnetic Nanoparticles. *Adv. Mater.* 2012, 24 (14), 1868-1872.
77. Chen, G. Y.; Pang, D. W. P.; Hwang, S. M.; Tuan, H. Y.; Hu, Y. C., A graphene-based platform for induced pluripotent stem cells culture and differentiation. *Biomaterials* 2012, 33 (2), 418-427.

78. Sun, X. M.; Liu, Z.; Welsher, K.; Robinson, J. T.; Goodwin, A.; Zaric, S.; Dai, H. J., Nano-Graphene Oxide for Cellular Imaging and Drug Delivery. *Nano Res* 2008, *1* (3), 203-212.
79. Liu, Z.; Robinson, J. T.; Sun, X. M.; Dai, H. J., PEGylated nanographene oxide for delivery of water-insoluble cancer drugs. *J. Am. Chem. Soc.* 2008, *130* (33), 10876-+.
80. Zhang, L. M.; Xia, J. G.; Zhao, Q. H.; Liu, L. W.; Zhang, Z. J., Functional Graphene Oxide as a Nanocarrier for Controlled Loading and Targeted Delivery of Mixed Anticancer Drugs. *Small* 2010, *6* (4), 537-544.
81. Zhou, L.; Wang, W.; Tang, J.; Zhou, J. H.; Jiang, H. J.; Shen, J., Graphene Oxide Noncovalent Photosensitizer and Its Anticancer Activity In Vitro. *Chem-Eur J* 2011, *17* (43), 12084-12091.
82. Zhang, W.; Guo, Z. Y.; Huang, D. Q.; Liu, Z. M.; Guo, X.; Zhong, H. Q., Synergistic effect of chemo-photothermal therapy using PEGylated graphene oxide. *Biomaterials* 2011, *32* (33), 8555-8561.
83. Hu, H. Q.; Yu, J. H.; Li, Y. Y.; Zhao, J.; Dong, H. Q., Engineering of a novel pluronic F127/graphene nanohybrid for pH responsive drug delivery. *Journal of Biomedical Materials Research Part A* 2012, *100A* (1), 141-148.
84. Pan, Y. Z.; Bao, H. Q.; Sahoo, N. G.; Wu, T. F.; Li, L., Water-Soluble Poly(N-isopropylacrylamide)-Graphene Sheets Synthesized via Click Chemistry for Drug Delivery. *Adv. Funct. Mater.* 2011, *21* (14), 2754-2763.
85. Feng, L. Z.; Zhang, S. A.; Liu, Z. A., Graphene based gene transfection. *Nanoscale* 2011, *3* (3), 1252-1257.
86. Scholz, W.; Boehm, H. P., Die Thermische Zersetzung Von Graphitoxyd. *Naturwissenschaften* 1964, *51* (7), 160.
87. Boehm, H. P.; Scholz, W., Der Verpuffungspunkt Des Graphitoxids. *Z. Anorg. Allg. Chem.* 1965, *335* (1-2), 74.

88. Boehm, H.; Clauss, A.; Fischer, G.; Hofmann, U. In *Surface properties of extremely thin graphite lamellae*, Proceedings of the fifth Conference on Carbon, Pergamon Press: 1962; pp 73-80.
89. Qiu, Y.; Guo, F.; Hurt, R.; Külaots, I., Explosive thermal reduction of graphene oxide-based materials: Mechanism and safety implications. *Carbon* 2014, 72 (0), 215–223.
90. Kim, F.; Luo, J. Y.; Cruz-Silva, R.; Cote, L. J.; Sohn, K.; Huang, J. X., Self-Propagating Domino-like Reactions in Oxidized Graphite. *Adv. Funct. Mater.* 2010, 20 (17), 2867-2873.
91. Botas, C.; Alvarez, P.; Blanco, C.; Santamaria, R.; Granda, M.; Gutierrez, M. D.; Rodriguez-Reinoso, F.; Menendez, R., Critical temperatures in the synthesis of graphene-like materials by thermal exfoliation-reduction of graphite oxide. *Carbon* 2013, 52, 476-485.
92. You, S. J.; Luzan, S. M.; Szabo, T.; Talyzin, A. V., Effect of synthesis method on solvation and exfoliation of graphite oxide. *Carbon* 2013, 52, 171-180.
93. Cataldo, F.; Compagnini, G.; D'Urso, L.; Palleschi, G.; Valentini, F.; Angelini, G.; Braun, T., Characterization of Graphene Nanoribbons from the Unzipping of MWCNTs. *Fuller Nanotub Car N* 2010, 18 (3), 261-272.
94. Jimenez, P. S. V., Thermal-Decomposition of Graphite Oxidation-Products Dsc Studies of Internal-Combustion of Graphite Oxide. *Mater. Res. Bull.* 1987, 22 (5), 601-608.
95. Mahajan, O. P.; Tomita, A.; Walker, P. L., Differential Scanning Calorimetry Studies on Coal .1. Pyrolysis in an Inert Atmosphere. *Fuel* 1976, 55 (1), 63-69.
96. Mohan, D.; Pittman, C. U.; Steele, P. H., Pyrolysis of wood/biomass for bio-oil: A critical review. *Energy Fuels* 2006, 20 (3), 848-889.
97. Antal, M. J.; Varhegyi, G., Cellulose Pyrolysis Kinetics - the Current State Knowledge. *Ind. Eng. Chem. Res.* 1995, 34 (3), 703-717.

98. Bradbury, A. G. W.; Sakai, Y.; Shafizadeh, F., A kinetic model for pyrolysis of cellulose. *J. Appl. Polym. Sci.* 1979, 23 (11), 3271-3280.
99. Kok, M. V.; Ozbas, E.; Karacan, O.; Hicyilmaz, C., Effect of particle size on coal pyrolysis. *J. Anal. Appl. Pyrolysis* 1998, 45 (2), 103-110.

**CHAPTER 2. GRAPHITE OXIDE
EXPLOSIVE THERMAL REDUCTION
AND SAFETY IMPLICATIONS FOR
LARGE SCALE STORAGE AND
PROCESSING**

This chapter is based on the published work by Y. Qiu, F. Guo, R. Hurt and I. Külaots, *Carbon* 2014.

2.1 Introduction

Graphene oxide and graphite oxide (GO) are promising precursors for large-scale manufacturing of graphene-based carbon materials. GO can be thermally or chemically treated to obtain reduced graphene oxide (rGO), which partially restores the electrical conductivity and hydrophobicity of pristine graphite for use as a 2D composite filler or conducting film. Processing of monolayer GO can lead to massive area loss (for example from 2600 to 40 m²g⁻¹) due to alignment and face-to-face stacking during GO deposition and drying ¹. Thermal exfoliation of bulk GO (graphite oxide) is attractive for large-scale production of GO-derived few-layer-graphene flakes ², or expanded graphene-based powders with high porosity and surface area for catalysis, separation, or gas storage applications. The heating of GO in inert gas is commonly called “thermal reduction” in the field because the main goal is to produce the reduced, graphene-like solid product, rGO. Strictly speaking, it is not a reduction since there is no external reducing agent, but is rather a chemical disproportionation, in which the original carbon atoms partition into reduced forms in solid rGO and oxidized forms that are primarily carbon oxide gas-phase byproducts (CO, CO₂). Thermal reduction of multilayer GO has been extensively investigated ³⁻⁹. High vacuum has been reported to support low temperature exfoliation (below about 300°C) by increasing the mechanical driving force for flake expansion, which is the difference between the internal (interstitial) pressure and the environmental pressure ^{6,9}. Porous materials from photothermal reduction of graphene oxide papers have been prepared by Mukherjee

et al.¹⁰ for lithium-ion battery applications. The presence of H₂ in the surrounding gas phase and pretreatment of GO with HCl have both been shown to enhance the thermal exfoliation of graphite oxide¹¹. Higher rates of thermal exfoliation are obtained by rapid release of the highly volatile HCl, which creates additional overpressure needed to successfully overcome van der Waals forces between GO sheets. In addition, hydrogen in gas environment during reduction can violently react to with –OH functionality in GO, and induce thermal decomposition¹¹. Thermal exfoliation in GO induces the decomposition of epoxides and hydroxyls, the rate of which competes with the diffusion rate of the reaction products CO₂ and CO decomposition. Successful thermal exfoliation is achieved when rate of decomposition of GO exceeds the rate of diffusion of gas products and needed threshold overpressure is built between individual GO layers¹². Increasing O/C ratio of GO⁴ increases GO decomposition rate and therefore, will enhance thermal exfoliation due the build up of larger gas volumes during reduction. For the successful thermal exfoliation overpressure is needed to overcome the van der Waals forces existing between the two adjacent GO layers¹².

It is well known that GO can be thermally unstable and should be regarded as an energetic material¹³⁻¹⁵. Nanoscale GO made from the oxidative unzipping of carbon nanotubes can also undergo explosive decomposition if heated in N₂ gas¹⁶. Kim et al.¹⁴ and Krishnan et al.¹⁵ report the spontaneous ignition of GO films in air under the influence of potassium residues that act as a catalyst for the carbon combustion reaction: $C + O_2 \Rightarrow CO/CO_2$, where “C” represents the rGO film. GO thermal reduction or disproportionation reaction is typically conducted in the absence of air, either in vacuum or inert gas blanket to protect the rGO product. Under these inert conditions there have been observations of a “popping” behavior -- rapid GO reduction with sudden onset in some experiments^{2,12}, but

no systematic studies of the factors that determine whether a given sample will exhibit the explosive or non-explosive mode.

We made the presumption that the self-initiating rapid reduction of GO with large-volume gas release would show features in common with other energetic materials (explosives and mono-propellants)¹⁷⁻¹⁹ and of some reactive chemicals that represent important safety concerns in industry²⁰⁻²², and must be understood and characterized for the safe handling of GO, especially at large scale. In the present study, we observe explosive decomposition during the thermal reduction of GO in inert gas environments leading to laboratory equipment damage, which occurs for some GO samples, but not others. We systematically explore the thermochemistry and mechanism of this energetic behavior and discuss its implications for safe GO processing and scale-up.

2.2 Materials and methods

2.2.1 Materials

SP-1 graphite powder was purchased from Bay Carbon (Bay City, MI). K₂S₂O₈, KMnO₄, reduced glutathione (GSH), ascorbic acid, hydrazine, H₃PO₄ and Acetone were purchased from Sigma Chemicals Co (St. Louis, MO). Trolox (6-Hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid), KOH, NaOH, Ca(OH)₂, NaNO₃, P₂O₅, H₂O₂, HCl and concentrated H₂SO₄ (98 %) were purchased from Fisher Scientific Inc. (Pittsburgh, PA).

GO synthesis was achieved through the modified Hummers method with pre-oxidation and two-step acid-acetone wash. Pre-oxidation was performed through mixing 14 g Graphite powder with 10 g K₂S₂O₈ and 10 g P₂O₅ in hot 98% H₂SO₄ and keeping stirring

at 80 °C for 5 hours. After filtration, the collected powder sample was washed with DI water and dried overnight. The pre-oxidized powder was then oxidized by a mixture containing 10 g NaNO₃, 70 g KMnO₄ in 98% H₂SO₄ at temperatures below 40 °C. After 3 hours stirring unreacted KMnO₄ is then neutralized by 30% H₂O₂. The sample was then purified by a two-step HCl-acetone wash to remove the salt byproducts ²³. The product was dried under ambient atmosphere in dark. The raw GO product was stored in the form of a lump, bulk solid, or GO cake. The aqueous suspensions of GO can be derived through dispersing 1.0 g GO in 500 mL nanopure water.

2.2.2 Methods

The GO multilayer films were prepared by pipetting 2 mg ml⁻¹ GO solution onto clean polystyrene substrate and left to dry at room temperature overnight (see procedure schematic in supporting information section of manuscript Fig.S1a). Each day the thickness of the existing GO film was increased by pipetting fresh 2 mgml⁻¹ GO solution on top of the previously dried film until the desired cycle number (cycle corresponds to each day) GO drop-cast film thickness was achieved. After drying was complete the x-cycle number GO film was peeled off from the polystyrene surface [see GO films in Figures S1b and S1c]. The total number of graphene oxide layers in each film can estimated from:

$$N = (m/A)/\rho\delta \quad (2.1)$$

where ρ is the material density, δ the interlayer spacing, m mass of GO and A the area of GO sample used.

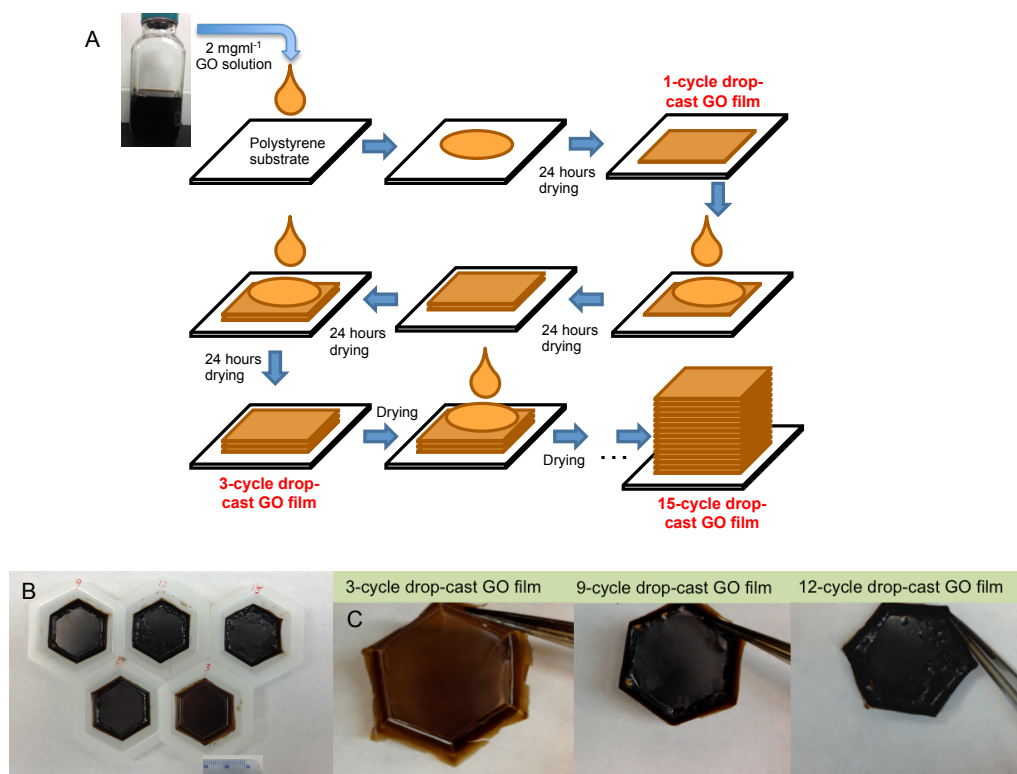


Figure 2.1 GO film preparation.

The GO multilayer films were prepared by pipetting 2 mgml⁻¹ GO solutions onto clean polystyrene substrate and left to dry at room temperature overnight. Each day the thickness of the existing GO film was increased by pipetting fresh 2 mgml⁻¹ GO solution on top of the previously dried film until the desired cycle number (each cycle corresponds to each day) GO drop-cast film thickness was achieved (A). The film thickness grows day by day (B). The dried x-cycle number GO film was peeled off from the polystyrene surface before conducting thermal analysis (C).

Drop-cast GO film and GO cake reduction experiments were performed in a differential scanning calorimeter (DSC) model 2910 from TA Instruments and thermogravimetric apparatus model 951 from TA Instruments in nitrogen gas flow. The heating rates applied were 10 to 50 Kmin⁻¹ and the maximum thermal reduction temperature 250°C. The surface morphology and the chemical composition of the rGO samples were characterized by field emission SEM with energy dispersive X-ray analysis (SEM-EDX). The metals content of GO cake samples was characterized by inductively coupled plasma emission spectroscopy (ICP-ES). The 77K N₂ and 273K CO₂ adsorption isotherms were recorded applying

Autosorb-1 instrument from Quantachrome, Inc. The BET surface areas were calculated from N₂ adsorption isotherms using relative pressures ranging from 0.05 to 0.30, if nearly perfect linear BET correlations were observed. The BET surface area was recorded when linear correlation factor at least $R^2 = 0.9999$ was achieved. Pore size distributions (PSD) were calculated applying non-local density functional theory (NLDFT) slit pore model ²⁴⁻²⁵.

2.3 Results and Discussion

2.3.1 Explosive thermal reduction of GO

During investigating GO thermal reduction, we occasionally observed violent decomposition events in the TGA in inert gas that resulted in loss of sample from the pan, failure of the experiment, and coating of very fine powder product throughout the interior of the device. The event was reproducible. More dramatically we observed an explosive decomposition in the DSC leading to interruption and failure of the experiment as well as DSC cell damage as shown in Figure 2.2A and Figure 2.2B. The metal DSC hermetic sample pan was torn into two parts, the reference pan was displaced and graphene-derived powder was spread throughout the cell. Images from the TGA experiment are also presented in Figure 2.2C (before micro-explosion) and in Figure 2.2D (after micro-explosion). In other reduction experiments in TGA and DSC, the integrity of the solid GO body was preserved. We hypothesized this damage was the result of a thermal runaway reaction driven by the exothermicity of GO reduction. We had not appreciated the potential for equipment damage when processing small amounts of GO, and were intrigued that some samples behaved energetically in inert gas, while others did not.

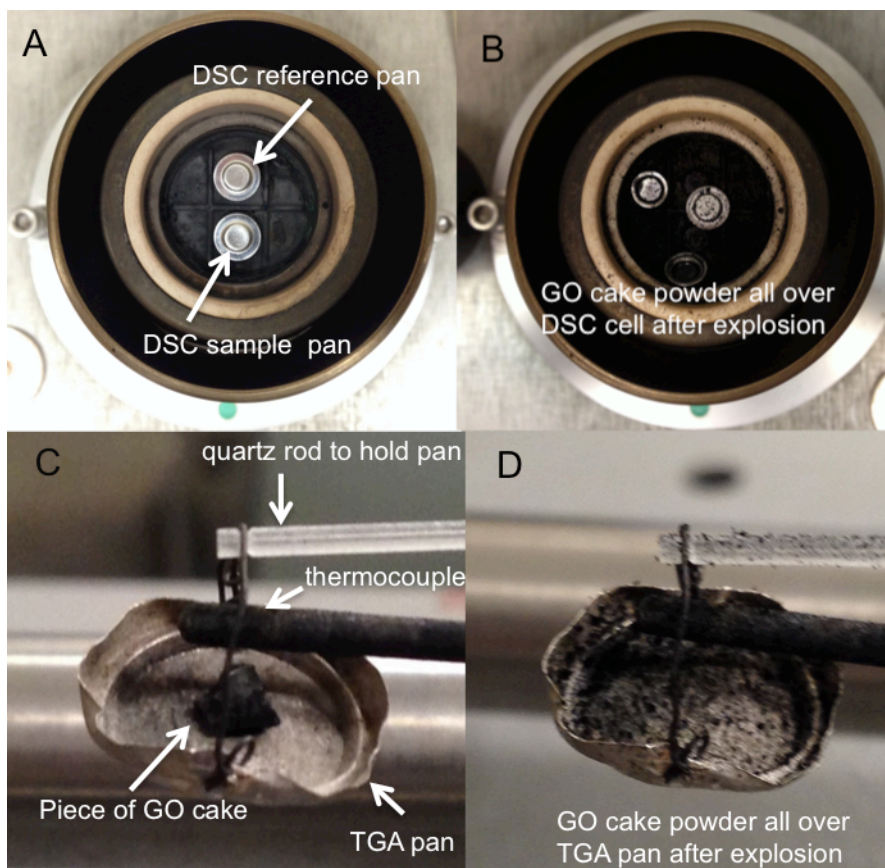


Figure 2.2 Images of DSC cell and TGA sample pan before and after micro-explosions.

Initially, approximately 2 mg of GO cake was placed into aluminum hermetic sample pan. Upper images show the hermetic pans before (A) and after (B) explosion. Note, after micro-explosion the hermetic pan is torn into two parts - the lid and the bottom parts, and both the sample and reference pan are misplaced. Black colored fine GO cake powder covers most of the cell. TGA sample pan images before (C) and after (D) explosion with 3.6 mg GO cake.

We therefore investigated the thermochemistry of GO reduction by employing uniform multilayer films of controlled thickness in DSC (see example thermogram shown in Figure 2.3A) and in TGA (see example TGA curve on Figure 2.3B and Figure 2.3C.). Thermal reduction of 15-cycle drop-cast film leads to a large exothermic peak in DSC, with decomposition ΔH of approximately $-1,680 \text{ Jg}^{-1}$. The onset temperature for the exothermic transition occurred near 150°C . This decomposition enthalpy ($1,680 \text{ Jg}^{-1}$) corresponds to an estimated adiabatic final temperature of $1,240^\circ\text{C}$, calculated for the case where the onset

temperature is 150°C and the product distribution is 40 wt% gas (as CO₂) and 60 wt% carbonaceous (as graphite) product. Adiabatic final temperature T_a was estimated from:

$$\Delta H = - \int_{T_o}^{T_a} C_p dT \quad (2.2)$$

where T_o is onset temperature, ΔH decomposition enthalpy as obtained experimentally with DSC, C_p specific heat capacities of the final products of CO₂ (polynomial applied) and of pure graphite.

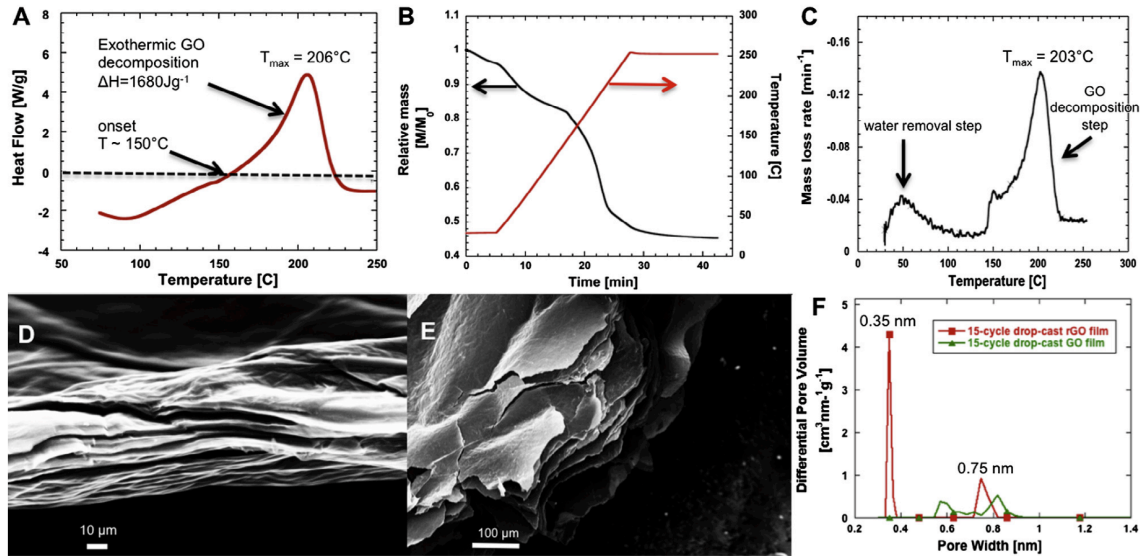


Figure 2.3 Thermal analysis of the GO reduction process and pore structure of the rGO product.

(A) DSC thermogram of 15-cycle drop-cast GO film ($1.1 \cdot 10^5$ GO layers) in 50 ml/min N₂ flow; heating rate 10 K/min, (B) TGA curve of the same 15-cycle drop-cast GO film in 100 ml/min N₂ flow; heating rate 10 K/min, (C) Differential TGA curve of the same GO film, (D) and (E) SEM images of 9-cycle drop-cast GO film ($6 \cdot 10^4$ GO layers) and 15-cycle drop-cast GO film ($1.1 \cdot 10^5$ GO layers). (F) 15-Cycle drop-cast GO and rGO film CO₂ isotherm NLDFT pore size distribution applying slit pore model.

In the DSC thermograph of GO undergoing non-explosive thermal reduction as shown in Figure 2.3A, the heat evolution was continuous and smooth and the reaction of GO in this film form did not produce a violent micro-explosion or sample pan damage. We hypothesize

that the thin film structure allow sufficiently fast heat transfer to prevent local temperature rise and thermal runaway, and allow sufficiently fast mass transfer of gas phase products to prevent internal pressure build-up and rGO film disintegration. Similar behaviour is seen when the 15-cycle drop-cast GO film was tested in TGA in inert gas environment. The GO film reduction does produce a porous rGO product, but does not explode (see rGO films on SEM images in Figure 2.3D and Figure 2.3E). The total mass loss in our GO decomposition is approximately 40 wt-% (not including water, see TGA example curve in Fig.1b), suggesting that our GO film has relatively high O/C ratio (30 wt-% reported in ^{2, 12}). Table 2.1 summarizes the DSC results for a number of experiments using GO samples of different form (cake or film) and mass. The temperatures at the peak of the DSC exotherm reported in Table 2.1 for all GO films are within the reported decomposition range for oxygen-containing functional groups on GO. As the drop-cast GO film thickness increases (while keeping the lateral area the same), the total energy released during GO decomposition increases linearly, and the correlation is independent of heating rates tested 10 or 50 K/min. The specific exothermic GO decomposition heat released is quite significant and is reproducible in the range 1400 to 1650 Joules per gram of GO, which is similar to the 1500 J/g reported by McAllister et al. ¹².

Krishnan et al. ¹⁵ reports a significantly higher exothermic decomposition enthalpy of 6 – 8 kJ/g, which may be due to differences in synthesis or to aging effects ²⁶, which require more study. All thermochemical data in Table 1 originate from the same original GO synthesis batch and none of these uniform multilayer films underwent explosive decomposition in this set of experiments, so the calorimetry results are quantitatively reliable. The FTIR scan results of 15-cycle drop-cast GO film and the same rGO film are shown on Figure S3c. As seen GO film investigated has wide hydroxyl peak (ranging between 2900-3600 cm⁻¹), together with epoxides (962 cm⁻¹), ethers (1035 cm⁻¹) and

carboxyls (1724 cm^{-1}). The 1619 cm^{-1} peak is related to vibrations from the unfunctionalized graphenic domains of GO. During thermal exfoliation in inert gas most of the oxides are removed when heated to 250°C . This result is consistent what has been as seen by others ^{2, 12}. Our SEM-EDX results give an average C/O atomic ratio of ~ 1.8 in our GO samples.

Table 2.1 Measured thermochemical parameters of graphene oxide reduction.

GO film drop-cast cycle number	GO layer number (estimated from GO mass)	Heating Rate, 10 K min^{-1}				Heating Rate, 50 K min^{-1}			
		GO [mg]	T_{max} [$^{\circ}\text{C}$]	ΔH [J/g]	Total heat [J]	GO [mg]	T_{max} [$^{\circ}\text{C}$]	ΔH [J/g]	Total heat [J]
6	3E+4	0.334	202	1568	0.524	0.463	230	1394	0.645
9	6E+4	0.648	200	1659	1.075	0.684	228	1459	0.998
12	9E+4	1.046	202	1639	1.714	1.064	227	1509	1.606
15	1.1E+5	1.242	206	1680	2.087	1.306	227	1457	1.903
GO cake	2.9E+5	3.250	190	1463	4.77	N/A	N/A	N/A	N/A

2.3.2 Morphology and pore structure of the rGO products

The surface area and porosity of the rGO samples listed in Table 2.1 were investigated both with N_2 and CO_2 probes. The N_2 and CO_2 adsorption analysis for BET surface area and PSD revealed that GO films prior to reduction have very low surface areas, similar to those reported ^{1, 6, 27}. For example, 3 and 6 cycle drop-cast GO film N_2 BET surface areas are 0.25 and $0.21\text{ m}^2\text{ g}^{-1}$, respectively and the corresponding thermal reduced samples have only slightly higher surface areas around $0.85\text{ m}^2\text{ g}^{-1}$. Therefore, low temperature, low heating

rate, and atmospheric pressure thermal reduction of thin GO films leads to relatively non-porous rGO. When a thicker film (15-cycle drop-cast) is reduced in DSC (SEM image shown on Figure 2.3E) we obtain much higher values: an N_2 surface area of $171 \text{ m}^2\text{g}^{-1}$, and a CO_2 BET surface area as large as $1471 \text{ m}^2\text{g}^{-1}$. To our knowledge this is the largest surface area reported for thermally reduced GO films. As the pore size distribution shows (Figure 2.3F) most of the regained surface area originates from super-micropores – those with pore sizes $\sim 0.35 \text{ nm}$. This is the micro-pore size range that can show molecular sieving behavior and allow access to CO_2 at 273 K , but not N_2 at 77 K due to activated diffusion processes ²⁸.

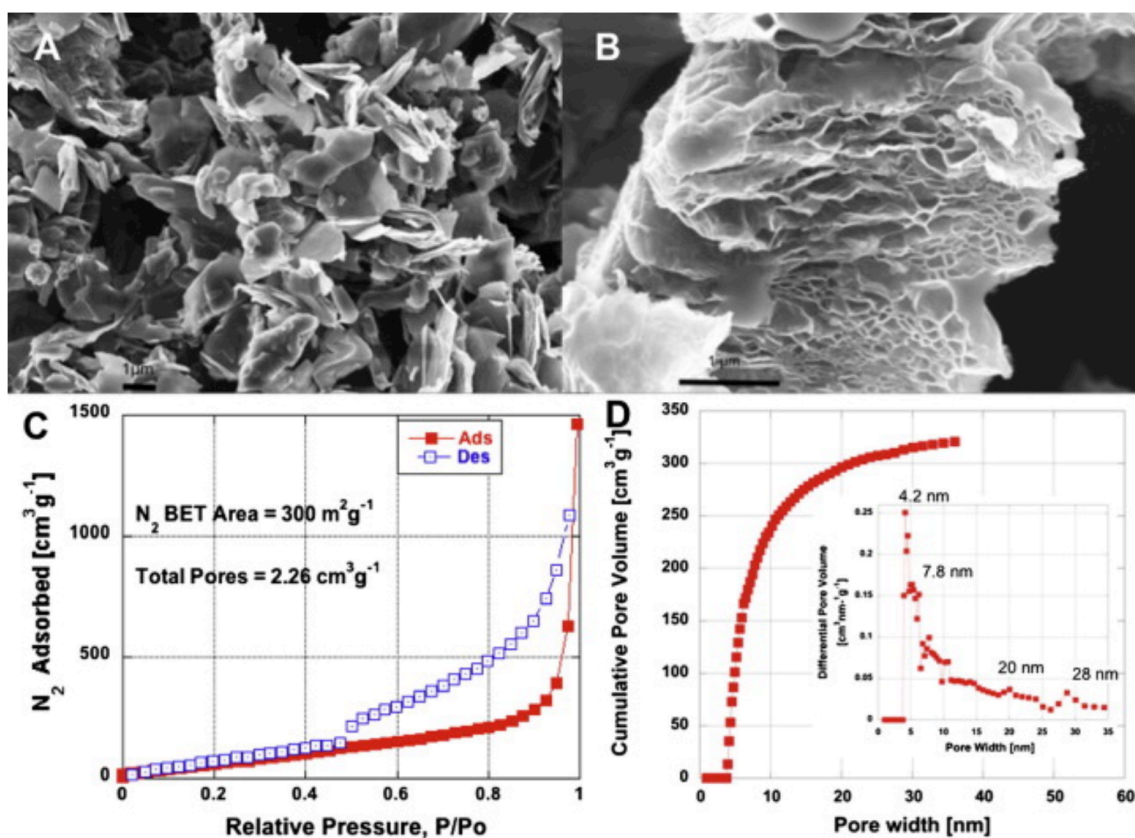


Figure 2.4 Morphology and pore structure of powder produced by explosive thermal reduction of GO cake.

SEM images (A and B), N_2 vapor adsorption isotherm (C), N_2 isotherm NLDFT and slit pore distribution (D).

Returning to the forensic investigation of why certain GO samples explode during thermal reduction, we examined the rGO powder product obtained after the micro-explosions of the GO cake in the DSC. Powders from the GO cake reduction were carefully removed from DSC cell (Figure 2.2B) and from the TGA pan (Figure 2.2D), and imaged in SEM. In Figure 2.4A the individual rGO particles are seen to have a highly irregular shape and with a lateral dimension from 1 to 3 μm . The SEM image on Figure 2.4B reveals that the powder shown in Figure 2.4A is multi-layer rGO platelet. Figure 2.4C and Figure 2.4D show the N_2 adsorption isotherm of the same rGO powder obtained after GO cake micro-explosion together with the pore size distribution (PSD) applying the non-local density function theory (NLDFT) slit pore model²⁴⁻²⁵. The BET N_2 surface area of the GO cake is only 2 m^2g^{-1} , while the exploded rGO cake product N_2 BET surface area is 300 m^2g^{-1} , implying a mean apparent layer number per stack of 8-9 using the scaling law $\text{S.A} \sim 2600N^{-1}$ ¹. The CO_2 BET surface area of GO cake is in the same magnitude as sensed with N_2 , but somewhat larger at 502 m^2g^{-1} , likely due to accessible internal super-microporosity. Interestingly, the rGO powder surface area is much smaller than the CO_2 BET surface area of non-exploded 15-cycle drop-cast rGO film as reported above 1471 m^2g^{-1} . This is an interesting finding, as it suggests that the micro-explosion is not necessarily desirable for achieving the highest possible surface area. The lower surface area in the exploded sample may be due to early release of the internal pressure when the film undergoes multipoint mechanical fracture partway through the gas evolution process. Samples that evolve gas quickly but not fast enough to explosively fracture likely develop the highest internal pressures to overcome interlayer van der Waals forces.

The PSD of GO cake powder (Figure 2.4D insert) reveals an interesting periodicity, which is similar to pore size periodicity reported in¹. This type of periodicity can be evidenced when investigating porosity of solid few-layer GO (FLGO) when each particle is considered as

thick platelet, which stay mostly non-porous and have relatively uniform thickness. The primary porosity generated in this type of GO system originates from the external surfaces of the multilayer rGO plates. A close examination of the PSD presented on Figure 2.4A insert suggests that the rGO plate thickness can be approximated to be 4 nm. Therefore suggesting that each rGO plate would include approximately 11 individual rGO sheet layers. From this layer number BET area is calculated to be $250 \text{ m}^2\text{g}^{-1}$ (applying power-law, $S.A = 2600N^{-1}$ as shown in ¹). This area result is in reasonable agreement with the measured $300 \text{ m}^2\text{g}^{-1} \text{ N}_2$ BET surface area.

2.3.3 Factors influencing the thermal reaction mode

2.3.3.1 Water

A series of control experiments were made to understand how the explosive mode reduction happens and its dependence on GO sample properties. Figure 2.5 shows a DSC thermogram of GO thermal reduction for an 8 mg cake sample in a sealed aluminum hermetic pan in inert gas is shown in Figure 2.5A. This is one of the GO cake experiments that produced explosive decomposition. The DSC thermogram on Figure 2.5A shows a very clear endothermic water peak with the maximum occurring at 126°C . The water removed at this temperature is likely bound water trapped between individual GO sheets by hydrogen bonding. The free external water is removed at around 50°C (see differential TGA curve Figure 2.3C), which DSC thermogram on Figure 2.5A does not show. As seen in thermogram Figure 2.5A the smooth endothermic water peak is followed by a GO thermal reduction exotherm, and in this case the peak is asymmetric and truncated with a smooth rise on the low-temperature side and an abrupt cliff on the high-temperature side. Throughout our DSC

experiments, this type of truncated exothermic peak correlates with the presence of the explosive event. The GO micro-explosion is preceded by a slight inflection in the DSC curve caused by local temperature rise (see arrow Figure 2.5A) that signals the runaway reaction, followed by a peak and abrupt decay as the sample pan loses contact with the DSC sensor. The onset temperature of the GO runaway reaction in this experiment is about $\sim 155^{\circ}\text{C}$ and the peak temperature is $\sim 171^{\circ}\text{C}$. We initially thought that interstitial water might be responsible for the violent GO micro-explosion during reduction in DSC by providing additional gas volume to increase internal interstitial pressure.

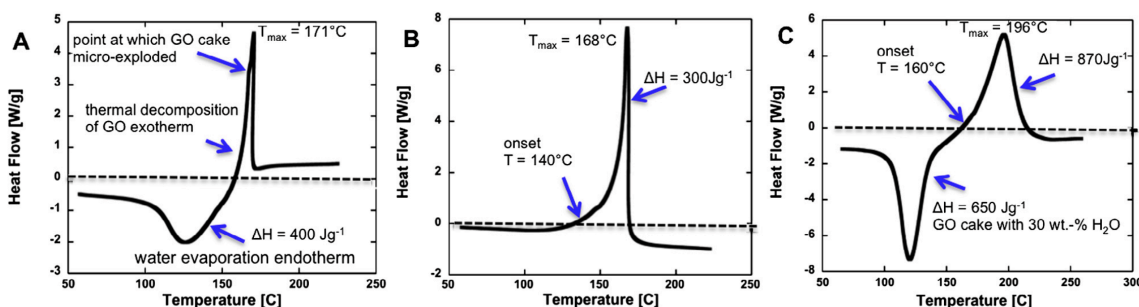


Figure 2.5 Effect of interstitial water on the mode of GO thermal reduction (explosive vs. non-explosive)-DSC thermograms

(A) GO cake which exploded during thermal reduction in N_2 gas, (B) 24 h low temperature oven dried GO cake (note GO cake still explodes after drying as indicated by the truncated asymmetric exotherm), (C) humidified GO cake DSC thermogram (note large water endotherm and smooth exotherm indicating no explosion). Interstitial water does not cause or promote the explosive mode of GO reduction.

In order to test the hypothesis that water is responsible of these micro-explosions, we oven-dried the GO cake sample that was known to explode (DSC thermogram shown on Figure 2.5A) overnight at relative low temperatures ($\sim 80^{\circ}\text{C}$) and repeated the DSC experiment in inert gas environment with heating rate 10 Kmin^{-1} . Figure 2.5B shows the oven-dried GO cake thermogram, which now is missing the endothermic water peak, but still ended with micro-explosion. Since the water endothermic peak is missing, one can easily determine the onset temperature of the GO thermal reduction as 140°C . In order to

further investigate the possible role of water in GO explosive reduction we humidified one of the GO cake samples that was known not to explode in a fully saturated water vapor environment at ambient temperature overnight. The thermogram in Figure 2.5C shows a very large endotherm, but the experiment did not end with the micro-explosion. One can note the large water endothermic peak, which specific heat determined from thermogram and the mass of water removed closely matched with the latent heat of water of evaporation 2,260 J/g at ambient pressure. The thermal reduction onset temperature is now at approximately 160°C, which is approximately ~20°C higher temperature if compared to GO cake onset temperature provided on Figure 2.5B. Note the smoothness of exothermic GO thermal reduction peak. The heat and mass generated during reduction has time to dissipate, and therefore the sample did not explode. These control experiments clearly show that water is not necessary for the explosive reduction of GO and does not promote the explosive mode of reduction. GO explosive decomposition is occurring in the dry state.

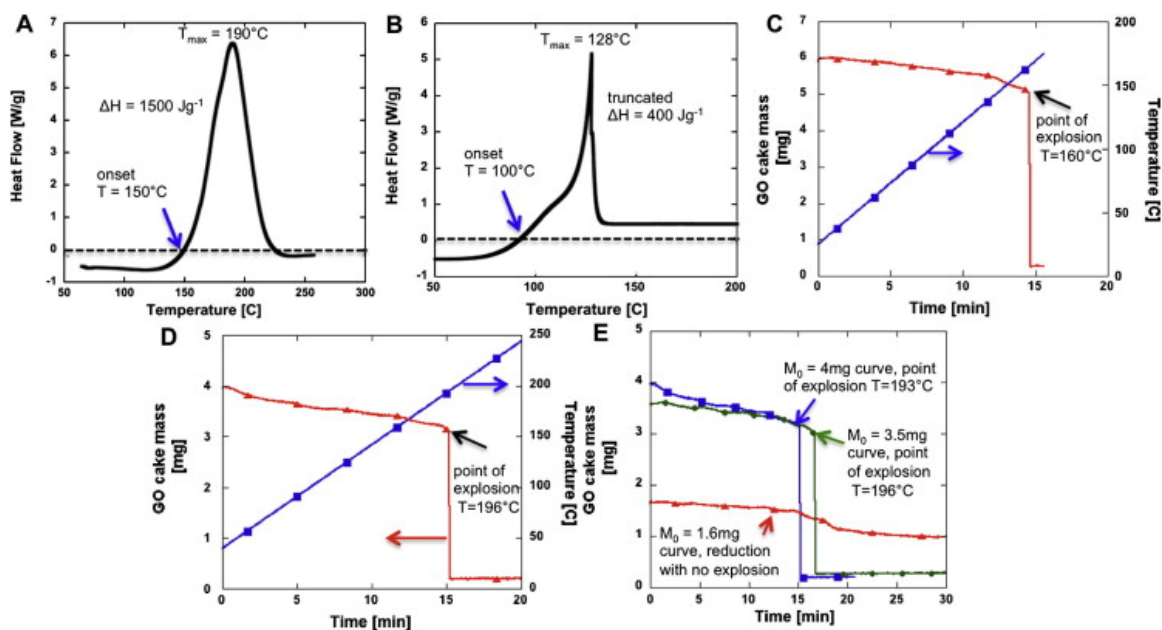


Figure 2.6 Role of potassium hydroxide immersion, residual potassium content (K), and total sample mass in the explosive mode of GO reduction.

(A) DSC thermogram of GO cake that did not explode, (B) DSC thermogram of the same cake immersed to 1 wt.% KOH solution and drying (note sharp exotherm which indicates explosive reduction). (C) and (D) TGA curves of two different GO cakes (C) GO cake which has 33-fold higher K content than the GO cake thermogram shown in (D). Note the K effect to explosion temperature, sample with high-K (C) explodes at 36 °C lower temperature compared to low-K GO sample in (D). (E) shows TGA curves of low-K content GO varying initial sample mass in thermal reduction: 4 mg, 3.5 mg and 1.6 mg. Sample with 1.6 mg does not explode, while the larger mass GO samples do.

2.3.3.2 Impurities

Further control experiments were conducted to see if explosive reduction is related to impurities. GO synthesis involves potassium permanganate KMnO_4 and samples may contain residual potassium ¹⁴⁻¹⁵. Potassium has been reported to cause GO ignition through catalysis of the combustion reaction ¹⁴⁻¹⁵, but there is no air in the current experiments, so combustion catalysis is not a possible mechanism. As a control experiment we used a GO cake sample that did not explode in the DSC (see smooth thermogram in Figure 2.6A) and immersed it into 1 wt-% KOH solution and repeated the DSC experiment (10 Kmin^{-1} in N_2 gas). The resulting DSC thermogram is shown in Figure 2.6B. There is no water endotherm present, and the cake did explode, indicating a promoting effect of potassium or hydroxide. In addition, the exotherm onset temperature is about 50°C lower than the undoped GO cake sample, which somewhat larger KOH treatment effect than 20°C lower onset temperature observed elsewhere ¹⁵. We conducted SEM EDX and ICP-AES experiments on two of the as-produced samples, only one of which was observed to undergo explosive reduction. The energetic sample had 8-fold higher potassium content (0.08 atom-%) than the non-energetic sample (0.01 atom-%) by EDX. A more accurate ICP-AES analysis showed potassium contents of 0.0436 ± 0.0017 wt.-% for the energetic sample and 0.0013 ± 0.0003 wt.-% for the non-energetic sample. Together these results suggest that potassium may play a role in promoting the explosive decomposition of GO. In addition to potassium's well-known behavior as a catalyst for carbon combustion, $\text{C} + \text{O}_2 \Rightarrow \text{CO/CO}_2$ ¹⁴, it has also been reported be catalysts for the pyrolysis of biomass and other oxygenated organic compounds ²⁹⁻³². Addition of

potassium to cellulosic material has been reported to increase gas and char (solid) yields, while reducing the yield of higher molecular weight volatiles (tars) ^{29, 31-32}. Potassium has been reported to increase CO and CO₂ yield ³⁰ or rate of evolution ²⁹ and to reduce the pyrolysis activation energy by 50 kJmol⁻¹ and lower the main pyrolysis temperature by 41-67°C ²⁹, which is very similar to the difference in onset temperatures seen in our GO thermal experiments (see Figure 2.6A and 2.6B). The molecular mechanism of K-catalysis of pyrolysis is not fully understood, but has been proposed to involve K-promoted heterolytic ring opening and cracking reactions ²⁹ and deoxygenation reactions ³⁰. Alternatively, the catalytic effect of KOH immersion may be due to hydroxide, which can establish basic conditions in the interstitial water phase. Several studies have reported base-catalyzed GO deoxygenation ³³⁻³⁵, which may reduce the exotherm onset temperatures observed here. More work is needed to understand the effect of impurities and additives on the potential for GO explosive decomposition.

Potassium hydroxide addition promotes GO explosive decomposition and lowers the onset temperature, but it remains to be seen if it is *necessary* for GO explosive decomposition. We therefore conducted series of TGA experiments with varying the GO cake mass. Figure 2.6C shows TGA thermogram of a GO cake sample known to explode in the DSC under identical heating conditions. The sample undergoes abrupt total mass loss indicating explosive reduction at 162°C, which is similar to the ~167°C explosion temperature seen in the DSC. A low-K GO cake that did not explode when used in film form in the DSC was also studied (see Figure 2.6D). This sample does explode in the TGA, but at 196°C, which is 34°C higher than the high-K cake (Figure 2.6C). Therefore, GO KOH immersion or high potassium content is not required for explosive decomposition.

2.3.3.3 Sample Mass

Table 2.2 Decomposition enthalpies for graphene oxide and a selection of energetic materials and reactive industrial chemicals known to pose processing safety hazards associated with thermal runaway reactions.

Material/chemical	Chemical application	Decomposition enthalpy (J g ⁻¹)	Reference
Graphene oxide (GO)	Precursor for reduced GO	1400–1700	Current study
Benzoyl peroxide	Industrial chemical	1602	Severini, 1984 ²²
Cumene hydroperoxide	Industrial chemical	1219	Di Somma, 2008 ²¹
Ethyl oleate ozonide	Industrial chemical	684	Cataldo 2013 ²⁰
Trinitrotoluene (TNT)	Monopropellant and explosive	2305	Ksiazczak 2000 ¹⁸
Hydrazine	Monopropellant and explosive	2980–4906	Jang et al. 2012 ¹⁷
Nitrocellulose	Monopropellant and explosive	1900–2400	Sovizi et al. 2009 ¹⁹

Figure 2.6E shows that the explosive reduction of GO can be initiated by increasing GO cake mass. In other known energetic materials, thermal runaway reactions occur when decomposition reaction heat cannot be dissipated fast enough to the surroundings, and local temperatures within the sample rise. Decomposition reactions have significant activation energies, and the elevated local temperatures thus cause accelerated decomposition rates and further heating in uncontrolled fashion leading to rapid gas release and pressure waves associated with deflagration or detonation mechanisms¹⁷. The stability of energetic materials is typically mass dependent, as the rate of heat generation is proportional to L^3 (for 3D bodies), while heat loss is related to surface area ($\sim L^2$) and size dependent heat transfer coefficients. For bulk GO samples, large sample mass favors internal buildup of both heat and gaseous reduction products, leading to thermal runaway and explosive exfoliation. It is significant that the explosive reduction can occur for GO sample masses as small as a few milligrams.

2.3.4 Safety implications

Even slow heating of few milligram-sized graphene oxide samples in inert gas can lead to uncontrolled micro-explosions that show the characteristics of thermal runaway reactions. The thermal runaway reaction is caused by the exothermicity of GO decomposition ($\sim 1,600 \text{ J g}^{-1}$) coupled with heat and mass transfer limitations that become more severe with increasing sample mass. Table 2 shows that GO mass-specific enthalpy of decomposition associated with its thermal reduction is comparable with the decomposition enthalpies of known hazardous industrial chemicals such as benzoyl peroxide ²², cumene hydroperoxide ²¹, and some explosives or monopropellants such as hydrazine ¹⁷, nitrocellulose ¹⁹ and trinitrotoluene (TNT) ¹⁸. The micro-explosions observed here occur for sample masses as low as few milligrams and can damage laboratory equipment, but a potentially more significant concern is the large-scale storage, handling, and processing of bulk graphene oxide as GO-based technologies are piloted and commercialized. The hazards associated with large bulk GO samples include heat generation, fire, gas release and overpressure in vessels, and potential pressure or shock waves when GO undergoes explosive reduction in confined environments or in open air, especially when self heating and self-initiation causes the thermal runaway reaction to occur in an unanticipated way during storage and handling.

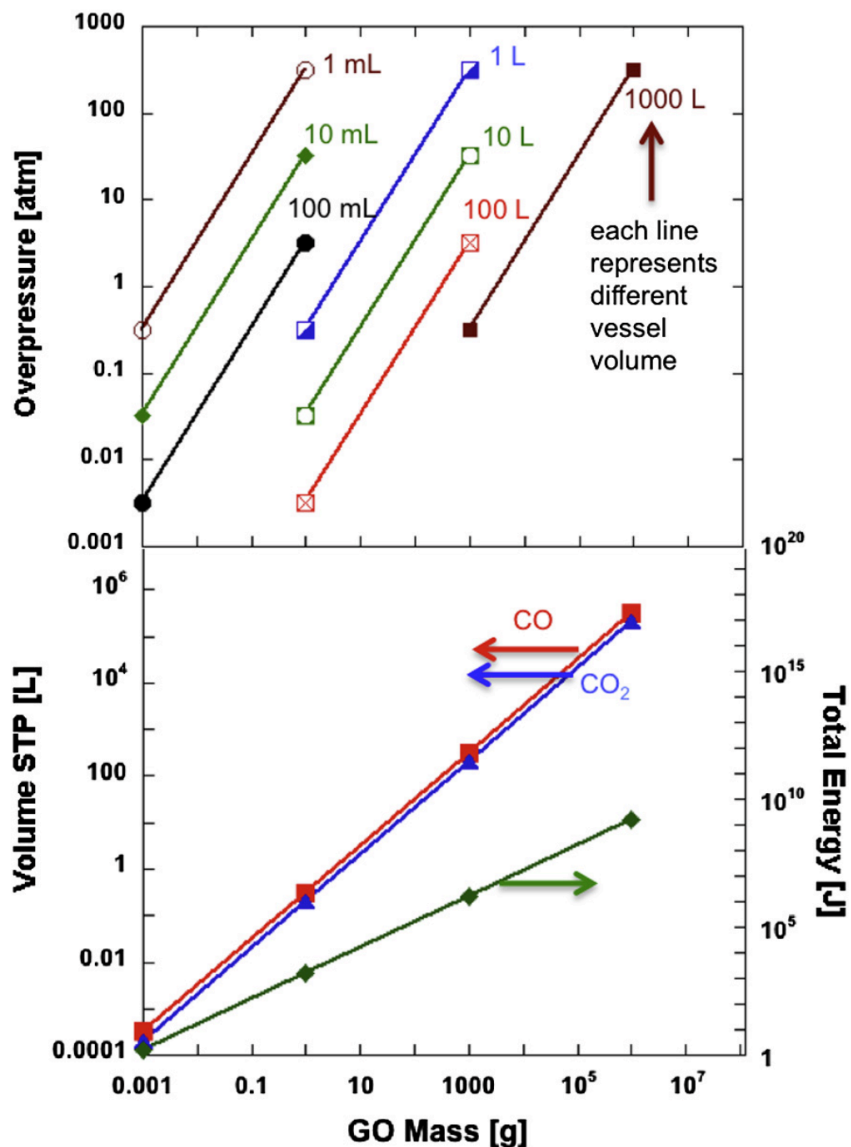


Figure 2.7 Safety implications of GO explosive reduction.

Bottom: total heat release and total evolved gas volume as a function of GO processing mass. The gas volume calculation was done for two limiting cases for the gas composition (pure CO_2 and pure CO), but the gas composition does not significantly affect the total gas volumes estimated. Top: Estimated overpressure following unanticipated GO decomposition in sealed processing or storage vessels of various sizes.

Figure 2.7 shows the results of calculations of heat release, volumetric gas release, and overpressure for a range of conditions and GO sample mass. The common x-axis is the mass of dry bulk GO, and y-axis volume of STP gas generated, total energy released (J) due to GO decomposition and overpressure (atm) created if sealed into closed containers with fixed

volume (varying container volumes from 1 ml to 100 liters shown curves on Figure 2.7 top). For example, if 1 gram of GO is used with the mass loss of roughly 40 % by weight, it will generate about 0.20 liters of gas and 1500 Joules of heat. Applying the ideal gas law and a chamber volume 1 ml 150°C temperature (typical GO runaway decomposition temperature), one can calculate the overpressure created during thermal reduction of 1 gram of GO to be 320 atmospheres. The mechanical stresses associated with overpressure may be augmented by pressure waves associated with deflagration or detonation processes. More work on the detailed kinetics and transport processes that govern GO explosive decomposition to understand the behavior as an energetic material.

2.4 Conclusions

The thermal reduction of graphene oxide samples in inert environments under slow heating can occur in either a controlled, continuous mode, or in an uncontrolled explosive mode. The explosive mode is a thermal runaway reaction driven by the exothermicity of GO reduction coupled with a threshold sample mass or minimum-dimension that retards heat and mass transfer allowing local temperature rise and internal pressure development. Understanding and managing the explosive mode of GO reduction is important for exfoliation efficiency, surface area development, and processing safety. Thermal reduction in the controlled (non-explosive) mode achieves an optimal CO₂ BET surface area as high as 1471 m²g⁻¹, which originates from pores approximately 0.35 nm in size. Explosive reduction leads to lower surface areas, as fracture causes early gas release that limits internal pressure development. The explosive mode of GO reduction in inert gas is not caused or promoted by the presence of interstitial water. Potassium hydroxide immersion lowers the

onset temperature for the GO reduction exotherm and increases the probability of the explosive mode. The intrinsic exothermicity of GO reduction ($\sim 1600 \text{ Jg}^{-1}$) is comparable to some hazardous industrial chemicals and explosives, and GO should thus be regarded as an energetic material that represents safety hazards during large-scale storage, handling, and processing. More work is needed to understand the kinetics and mechanisms of the explosive reduction process in order to fully characterize the hazards associated with large-scale GO processing and allow its safe development and commercialization.

2.5 References

1. Guo, F.; Creighton, M.; Chen, Y.; Hurt, R.; Külaots, L., Porous structures in stacked, crumpled and pillared graphene-based 3D materials. *Carbon* 2014, 66 (0), 476-484.
2. Zhang, C.; Lv, W.; Xie, X.; Tang, D.; Liu, C.; Yang, Q.-H., Towards low temperature thermal exfoliation of graphite oxide for graphene production. *Carbon* 2013, 62, 11-24.
3. Du, Q. L.; Zheng, M. B.; Zhang, L. F.; Wang, Y. W.; Chen, J. H.; Xue, L. P.; Dai, W. J.; Ji, G. B.; Cao, J. M., Preparation of functionalized graphene sheets by a low-temperature thermal exfoliation approach and their electrochemical supercapacitive behaviors. *Electrochim. Acta* 2010, 55 (12), 3897-3903.
4. Jin, M.; Jeong, H. K.; Kim, T. H.; So, K. P.; Cui, Y.; Yu, W. J.; Ra, E. J.; Lee, Y. H., Synthesis and systematic characterization of functionalized graphene sheets generated by thermal exfoliation at low temperature. *Journal of Physics D-Applied Physics* 2010, 43 (27).

5. Jung, I.; Dikin, D. A.; Piner, R. D.; Ruoff, R. S., Tunable electrical conductivity of individual graphene oxide sheets reduced at "low" temperatures. *Nano Lett.* 2008, 8 (12), 4283-4287.
6. Lv, W.; Tang, D. M.; He, Y. B.; You, C. H.; Shi, Z. Q.; Chen, X. C.; Chen, C. M.; Hou, P. X.; Liu, C.; Yang, Q. H., Low-Temperature Exfoliated Graphenes: Vacuum-Promoted Exfoliation and Electrochemical Energy Storage. *ACS nano* 2009, 3 (11), 3730-3736.
7. Schniepp, H. C.; Li, J. L.; McAllister, M. J.; Sai, H.; Herrera-Alonso, M.; Adamson, D. H.; Prud'homme, R. K.; Car, R.; Saville, D. A.; Aksay, I. A., Functionalized single graphene sheets derived from splitting graphite oxide. *J. Phys. Chem. B* 2006, 110 (17), 8535-8539.
8. Ye, J.; Zhang, H. Y.; Chen, Y. M.; Cheng, Z. D.; Hu, L.; Ran, Q. Y., Supercapacitors based on low-temperature partially exfoliated and reduced graphite oxide. *J. Power Sources* 2012, 212, 105-110.
9. Zhang, H. B.; Wang, J. W.; Yan, Q.; Zheng, W. G.; Chen, C.; Yu, Z. Z., Vacuum-assisted synthesis of graphene from thermal exfoliation and reduction of graphite oxide. *J. Mater. Chem.* 2011, 21 (14), 5392-5397.
10. Mukherjee, R.; Thomas, A. V.; Krishnamurthy, A.; Koratkar, N., Photothermally reduced graphene as high-power anodes for lithium-ion batteries. *ACS nano* 2012, 6 (9), 7867-7878.
11. Shen, B.; Lu, D. D.; Zhai, W. T.; Zheng, W. G., Synthesis of graphene by low-temperature exfoliation and reduction of graphite oxide under ambient atmosphere. *Journal of Materials Chemistry C* 2013, 1 (1), 50-53.
12. McAllister, M. J.; Li, J. L.; Adamson, D. H.; Schniepp, H. C.; Abdala, A. A.; Liu, J.; Herrera-Alonso, M.; Milius, D. L.; Car, R.; Prud'homme, R. K.; Aksay, I. A., Single sheet

functionalized graphene by oxidation and thermal expansion of graphite. *Chem. Mater.* 2007, 19 (18), 4396-4404.

13. Jimenez, P. S. V., Thermal-Decomposition of Graphite Oxidation-Products Dsc Studies of Internal-Combustion of Graphite Oxide. *Mater. Res. Bull.* 1987, 22 (5), 601-608.

14. Kim, F.; Luo, J. Y.; Cruz-Silva, R.; Cote, L. J.; Sohn, K.; Huang, J. X., Self-Propagating Domino-like Reactions in Oxidized Graphite. *Adv. Funct. Mater.* 2010, 20 (17), 2867-2873.

15. Krishnan, D.; Kim, F.; Luo, J. Y.; Cruz-Silva, R.; Cote, L. J.; Jang, H. D.; Huang, J. X., Energetic graphene oxide: Challenges and opportunities. *Nano Today* 2012, 7 (2), 137-152.

16. Cataldo, F.; Compagnini, G.; D'Urso, L.; Palleschi, G.; Valentini, F.; Angelini, G.; Braun, T., Characterization of Graphene Nanoribbons from the Unzipping of MWCNTs. *Fuller Nanotub Car N* 2010, 18 (3), 261-272.

17. Jang, I. J.; Shin, H. S.; Shin, N. R.; Kim, S. H.; Kim, S. K.; Yu, M. J.; Cho, S. J., Macroporous-mesoporous alumina supported iridium catalyst for hydrazine decomposition. *Catal. Today* 2012, 185 (1), 198-204.

18. Ksiazczak, A.; Ksiazczak, T., Influence of DSC measurement conditions on kinetic parameters of thermal decomposition of 2,4,6-trinitrotoluene. *J. Therm. Anal. Calorim.* 2000, 60 (1), 25-33.

19. Sovizi, M. R.; Hajimirsadeghi, S. S.; Naderizadeh, B., Effect of particle size on thermal decomposition of nitrocellulose. *J. Hazard. Mater.* 2009, 168 (2-3), 1134-1139.

20. Cataldo, F., Chemical and thermochemical aspects of the ozonolysis of ethyl oleate: decomposition enthalpy of ethyl oleate ozonide. *Chem. Phys. Lipids* 2013, 175, 41-49.

21. Di Somma, I.; Andreozzi, R.; Canterino, M.; Caprio, V.; Sanchirico, R., Thermal decomposition of cumene hydroperoxide: chemical and kinetic characterization. *AIChE J.* 2008, 54 (6), 1579-1584.

22. Severini, F.; Gallo, R., Differential scanning calorimetry study of thermal-decomposition of benzoyl peroxide and 2,2'-azobisisobutyronitrile mixtures. *J. Therm. Anal.* 1984, 29 (3), 561-566.
23. Kim, J.; Cote, L. J.; Kim, F.; Yuan, W.; Shull, K. R.; Huang, J. X., Graphene Oxide Sheets at Interfaces. *J. Am. Chem. Soc.* 2010, 132 (23), 8180-8186.
24. Lastoskie, C.; Gubbins, K. E.; Quirke, N., Pore-Size Heterogeneity and the Carbon Slit Pore - a Density-Functional Theory Model. *Langmuir* 1993, 9 (10), 2693-2702.
25. Ravikovitch, P. I.; Vishnyakov, A.; Russo, R.; Neimark, A. V., Unified approach to pore size characterization of microporous carbonaceous materials from N₂, Ar, and CO₂ adsorption isotherms. *Langmuir* 2000, 16 (5), 2311-2320.
26. Kim, S.; Zhou, S.; Hu, Y. K.; Acik, M.; Chabal, Y. J.; Berger, C.; de Heer, W.; Bongiorno, A.; Riedo, E., Room-temperature metastability of multilayer graphene oxide films. *Nat Mater* 2012, 11 (6), 544-549.
27. Yang, S. J.; Kim, T.; Jung, H.; Park, C. R., The effect of heating rate on porosity production during the low temperature reduction of graphite oxide. *Carbon* 2013, 53, 73-80.
28. Lozano-Castello, D.; Cazorla-Amoros, D.; Linares-Solano, A., Usefulness of CO₂ adsorption at 273 K for the characterization of porous carbons. *Carbon* 2004, 42 (7), 1233-1242.
29. Nowakowski, D. J.; Jones, J. M.; Brydson, R. M. D.; Ross, A. B., Potassium catalysis in the pyrolysis behaviour of short rotation willow coppice. *Fuel* 2007, 86 (15), 2389-2402.
30. Pan, W. P.; Richards, G. N., Influence of metal-ions on volatile products of pyrolysis of wood. *J. Anal. Appl. Pyrolysis* 1989, 16 (2), 117-126.
31. Raveendran, K.; Ganesh, A.; Khilar, K. C., Influence of mineral matter on biomass pyrolysis characteristics. *Fuel* 1995, 74 (12), 1812-1822.

32. Wang, Z. Y.; Wang, F.; Cao, J. Q.; Wang, J., Pyrolysis of pine wood in a slowly heating fixed-bed reactor: Potassium carbonate versus calcium hydroxide as a catalyst. *Fuel Process. Technol.* 2010, *91* (8), 942-950.
33. Dimiev, A. M.; Alemany, L. B.; Tour, J. M., Graphene oxide. Origin of acidity, its instability in water, and a new dynamic structural model. *ACS nano* 2013, *7* (1), 576-88.
34. Fan, X. B.; Peng, W. C.; Li, Y.; Li, X. Y.; Wang, S. L.; Zhang, G. L.; Zhang, F. B., Deoxygenation of Exfoliated Graphite Oxide under Alkaline Conditions: A Green Route to Graphene Preparation. *Adv. Mater.* 2008, *20* (23), 4490-4493.
35. Wang, F. T. T.; Chen, L.; Tian, C. J. J.; Meng, Y.; Wang, Z. G. G.; Zhang, R. Q. Q.; Jin, M. X. X.; Zhang, P.; Ding, D. J. J., Interactions Between Free Radicals and a Graphene Fragment: Physical Versus Chemical Bonding, Charge Transfer, and Deformation. *J. Comput. Chem.* 2011, *32* (15), 3264-3268.

**CHAPTER 3 KINETICS AND
THERMOCHEMISTRY OF GRAPHITE
OXIDE EXOTHERMIC THERMAL
REDUCTION**

3.1 Introduction

Graphite oxide (GO) is the bulk raw product of graphite oxidation with strong oxidants that intercalate the graphite crystal lattice and introduce oxygen functional groups to the basal plane. GO has long been recognized as a thermally unstable, energetic material ¹⁻¹². For example, Boehm et al in 1962 reported “when heated quickly to about 180°C it deflagrates with production of a very voluminous fluffy carbon”, which “consists of very thin carbon sheets” ⁴.

Isolation of the graphene monolayer in 2004 ¹³⁻¹⁵ has greatly increased interests in this old material, GO, first for its role as an intermediate in the chemical route to graphene, and more recently as a precursor for solution exfoliation into graphene oxide as an end product. Graphene oxide is a hydrophilic water-processable two-dimensional monolayer material with its own applications (e.g. in membranes ¹⁶, emulsifiers ¹⁷, hydrogels ¹⁸, aerogels ¹⁹ and sorbents ²⁰), some of which are distinct from the applications of pristine graphene. GO is also of great interest as a bulk starting material for controlled thermal exfoliation into multilayer rGO flakes used as composite fillers ²¹⁻²⁴. High-aspect-ratio flakes achieve low percolation thresholds, and are thus especially attractive in conductive composites ^{21, 25-26}.

The current strong focus on graphene applications ²⁷⁻³¹ together with the important role of GO as an intermediate in some processes, have now led to an intense interest in the large-scale manufacture and commercialization of bulk GO. With large-scale manufacturing also come scale-up issues, which include safety, and this motivates a much closer look at the behavior of GO as an energetic material under typical manufacturing and process conditions ⁵.

The thermal decomposition of GO yields large volumes of gaseous products, typically from 40 up to 60% per mass of the original sample mass depending on initial C:O ratio. The decomposition is highly exothermic and accompanied by self-heating, which for bulk samples above a critical size can lead to significant heat accumulation and thermal runaway reaction. If this occurs spontaneously in an unexpected or undesired processing or storage step, it can lead to gas release, vessel overpressure ^{5,10}, or ignition of the volatile gases with atmospheric oxygen and large-scale fire hazard ³².

This study addresses important unresolved issues that impact GO processing safety at large scale. First there is an unexplained large variation in GO decomposition exotherm onset temperatures. The onset temperature marks the practical start of the self-heating process and defines the maximum temperature for safe storage and processing. Many studies report onset temperatures in the range of 150-220°C ^{10-11, 33-34}, but our preliminary data also identifies samples with onset temperatures near 100°C, which coincides with typical condition used in common drying operations ^{8, 35-36}. What causes low GO decomposition onset temperature in some samples, and how can it be avoided? Secondly, there is a relatively large variation in the reported magnitudes of the exotherm ranging from 1000 to 8000 J g⁻¹ ^{7, 10, 32}. Also, to our knowledge there are no data on enthalpy of formation and combustion of GO. Thirdly, GO has been reported to undergo dynamic aging processes ³⁷⁻⁴¹, whose effect on thermal decomposition reactivity and heat release are unknown. Finally, GO is a complex material, and work is needed to understand the role of partial reduction, salt impurities ¹⁷, and suspension pH prior to drying in the thermal decomposition process.

We address each of these issues by measuring quantitative thermochemical and kinetic parameters for GO thermal decomposition. Those kinetics are then used in a computational

study of the sample sizes and environmental temperatures required to avoid self-heating and allow safe storage and processing that will be needed for successful nano-manufacturing of GO-based materials.

3.2 Materials and methods

3.2.1 Materials

GO was produced with the method as described in the last chapter. The desired mass of additives (KOH, KCl, ascorbate etc.) was weighed and transferred into a solution with fixed concentration by adding DI water. Then, the desired mass of solid GO was added into the premixed solution (with dry GO: additive mass ratio of 100:1). The mixture was agitated vigorously using a vortex mixer for approximately 30 seconds, after which, vials with GO plus salt solution were left uncapped in a 60°C oven overnight to dry. The resulting product is in the form of flaky powder.

3.2.2 Characterization

3.2.2.1 Morphology

The morphologies of all the materials used in this work were characterized with a LEO 1530 field-emission scanning electron microscope (SEM). The oxygen functional groups on the graphenic plane and edges were characterized using ATR accessory in Fourier Transform Infrared Spectroscopy (JASCO FT/IR-4100) and X-ray photoelectron spectroscopy (XPS) on Physical Electronics 5500 (PHI 5500) Multi-technique Surface Analyzer.

3.2.2.2 Thermal analysis

TA Instruments DSC-2910 Differential Scanning Calorimeter was applied to measure enthalpies of decomposition, and determine GO decomposition reaction onset temperatures. MT TGA/DSC-1 instrument was applied to test simultaneous mass loss and heat effects of GO. A heating rate of 10 K min⁻¹ was used in most experiments, except in the kinetics experiments, where it was systematically varied to determine activation energy and frequency factor. The enthalpy of combustion of GO was determined by using Parr Instruments model 1421 semi-micro oxygen bomb calorimeter.

3.2.3 Decomposition kinetics and thermochemistry analysis

3.2.3.1 Decomposition reaction kinetics

Decomposition reaction rate constant activation energy and frequency factor were defined from variable heating rate DSC data applying Kissinger equation ⁴²

$$\ln\left(\frac{\phi}{T_m^2}\right) = \ln\left(\frac{AR}{E_a}\right) - \frac{E_a}{RT_m} \quad (3.1)$$

where ϕ is heating rate; T_m , the temperature at which the heat flow reaches a maximum; A , frequency factor; E_a , activation energy. The empirical reaction order n was first estimated using Eq. (3.2) ⁴² and then verified by fitting isothermal DSC data with the kinetic model represented by Eq. (3.1) and (3.2).

$$n = 1.26 S^{0.5} \quad (3.2)$$

In Eq. (3.2), S is the curve shape index defined as the absolute value of the ratio of the slopes of tangents to the DSC curve at the two inflection points.

3.2.3.2 Self-heating analysis

Self-heating simulations were conducted applying the following energy balance equation

$$mC_p \frac{dT}{dt} = -m\Delta H \frac{dx}{dt} - hS(T - T_0) \quad (3.3)$$

where m is the mass of GO, C_p is the specific heat capacity, x is the mass fraction reacted, ΔH is the enthalpy of decomposition, h is the heat transfer coefficient to the local environment, T_0 is surrounding environment temperature (which was also taken to be the initial temperature of GO) and S is bulk GO external surface area, which is exposed to gas atmosphere. Adiabatic and non-adiabatic condition self-heating curves were computed applying numerical methods to Eq. (3.3).

3.2.3.3 Enthalpy of decomposition analysis

GO thermal reduction is essentially an organic disproportionation or decomposition reaction expressed as



where rGO is solid reduced graphite oxide product. The enthalpy of decomposition for the reaction above can be estimated if the GO and rGO enthalpies of formation are known. In this work, we used the group contribution method, proposed by J. Marrero and R. Gani⁴³ to obtain the GO enthalpy of formation for a variety of hypothetical GO organic structures. By introducing varying amounts of epoxides or hydroxyls to a polyaromatic (graphene) cluster with periodic boundaries, the quantitative contributions of these two types of groups to the GO enthalpy of formation is estimated. More details about these calculations can be found in electronic supplemental information section.

3.3 Results and discussion

3.3.1 GO thermal exfoliation in nitrogen and air atmospheres

Figure 3.1 illustrates the basic thermochemical behavior of GO upon slow heating at 10 K min⁻¹. In DSC under N₂ flow GO shows a small endotherm associated with water evaporation followed by a single exotherm corresponding to thermal decomposition with an onset temperature near 150°C and a total heat release of ~1600 J g⁻¹. When heated in air, a second exotherm onset temperature also appears at ~ 550°C, which is a typical onset temperature for the oxidation of disordered or defective carbons prepared at low temperature ⁴⁴⁻⁴⁵. Integration of the higher-temperature peak gives a reaction enthalpy of 3920 J g⁻¹-initial-GO. This result is in good agreement with the theoretical standard heat of reaction for the incomplete carbon (rGO) oxidation to CO: $C + 1/2O_2 \rightarrow CO$, which is 9200 J g⁻¹-C, and when renormalized to initial GO mass gives a value similar to the experimental result of 3900 J g⁻¹ initial-GO-solid. The renormalization is performed by using GO C:O atomic ratio 2:1, and assuming the decomposition products as rGO, CO and CO₂ with CO:CO₂ ratio of 1:2 in gas products (CO:CO₂ ratio based on study in Jung et al., 2009⁴⁶).

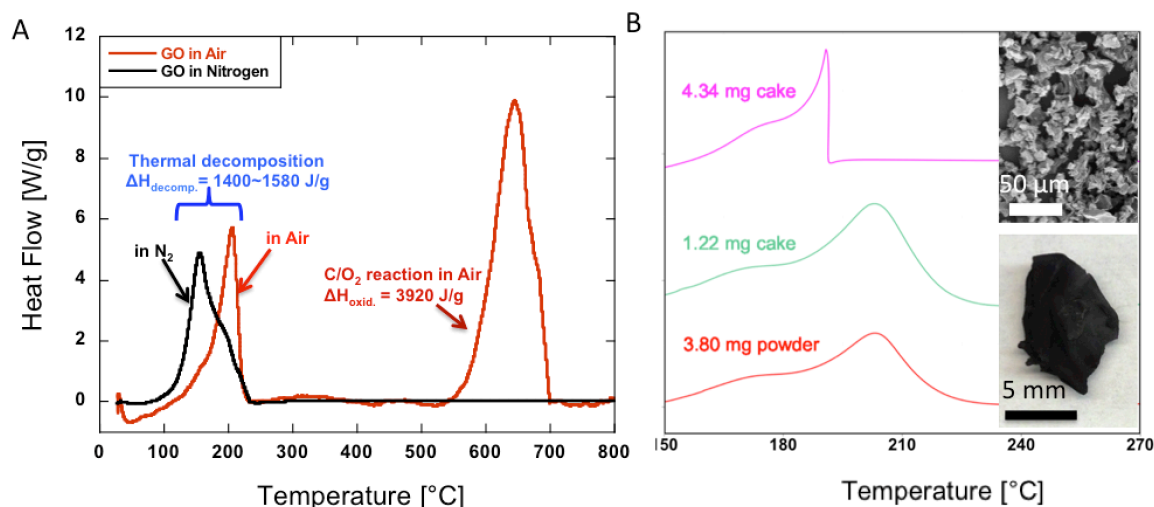


Figure 3.1 DSC thermograms of GO thermal reduction under different condition

(A) DSC thermograms of GO thermal reduction in nitrogen compared with the same sample reduction and oxidation in air. (B) The GO initial mass and solid form (powder versus single large piece) determine if the decomposition of GO occurs in explosive or non-explosive modes. The non-explosive thermal reduction produces a large rGO monolith similar to the starting form of the GO cake (lower inset image), while the explosive mode produces rGO in the form of micron-sized particles (upper inset image).

We have also measured the total enthalpy of combustion of GO using an oxygen bomb calorimeter. Our duplicate experiments yielded a GO heat of combustion of $14,100 \text{ J g}^{-1}$ initial-GO-solid, which is within a range of enthalpy of combustion for low calorie content solid fuels such as lignites, brown coals and biomasses. Based on our experimental measurements, theoretical predictions and literature review, it appears that the true decomposition enthalpy for GO is clearly in the range of $1000\text{--}2000 \text{ J g}^{-1}$, and the isolated reports of much higher values ($\sim 8000 \text{ J g}^{-1}$) may reflect studies carried out in air, which effectively report the sum of the decomposition enthalpy and further oxidation of either the char (rGO) or the gaseous decomposition products.

The smooth DSC heat decomposition curves in Figures 1A and 1B change to a truncated, asymmetric curve when larger sample size is used (see top curve on Figure 3.1B). This behavior is known to represent the explosive mode thermal decomposition, which occurs

when GO decomposition reaction rate exceeds mass and heat transfer rates leading to local self-heating and thermal runaway reaction ^{5, 10, 32}. Both the sample mass and sample form (powder vs. consolidated “cake”) affect this transition (Figure 3.1B). While we are interested in the explosive events, and in ways to avoid them during processing and storage, the remainder of this study is carried out with relatively small size samples to achieve the non-explosive mode decomposition, which is required to obtain the smooth non-truncated heat curves needed to accurately measure and report quantitative kinetic and thermochemical parameters.

3.3.2 Role of salt impurities and reducing agents in GO decomposition reaction

Figure 3.2 explores the role of various salt impurities and chemical additives on GO thermal behavior. Potassium is a common impurity as a residue of the potassium permanganate used in graphite oxidation, and has been reported to promote ignition of GO upon heating ³². Our previous study ⁵ reported that KOH addition reduced the onset temperature for GO thermal decomposition, and in some cases the initial portion of the exotherm extended down into the range of common drying operations (100-120°C). We have undertaken the study to understand the effect and the mechanism of KOH treatment, because such a low onset temperature raises special safety concerns. The new data in Figure 3.2A,B clearly show that the catalytic effect of KOH on GO decomposition is due to OH⁻ and not to K⁺. Hydroxyl ion has been previously reported to be a catalyst for GO reduction in aqueous suspension ⁴⁰, but this is the first report of its effect as a catalyst for GO thermal decomposition in dried samples. While K⁺ does not effect GO thermal decomposition, it does catalyze the subsequent carbon (rGO) combustion reaction occurring at much higher

temperatures (Figure 3.2C). Potassium addition reduces the onset temperature for $C+O_2 \rightarrow CO/CO_2$ by up to 200°C in Figure 3.2C, which is consistent with potassium's known role as an active char combustion catalyst⁴⁷⁻⁴⁹. These results indicate that when GO is heated in air, the presence of residual K^+ will not promote the original decomposition, but can increase the probability of subsequent ignition and rGO combustion with corresponding fire hazard³² (Figure 3.2E).

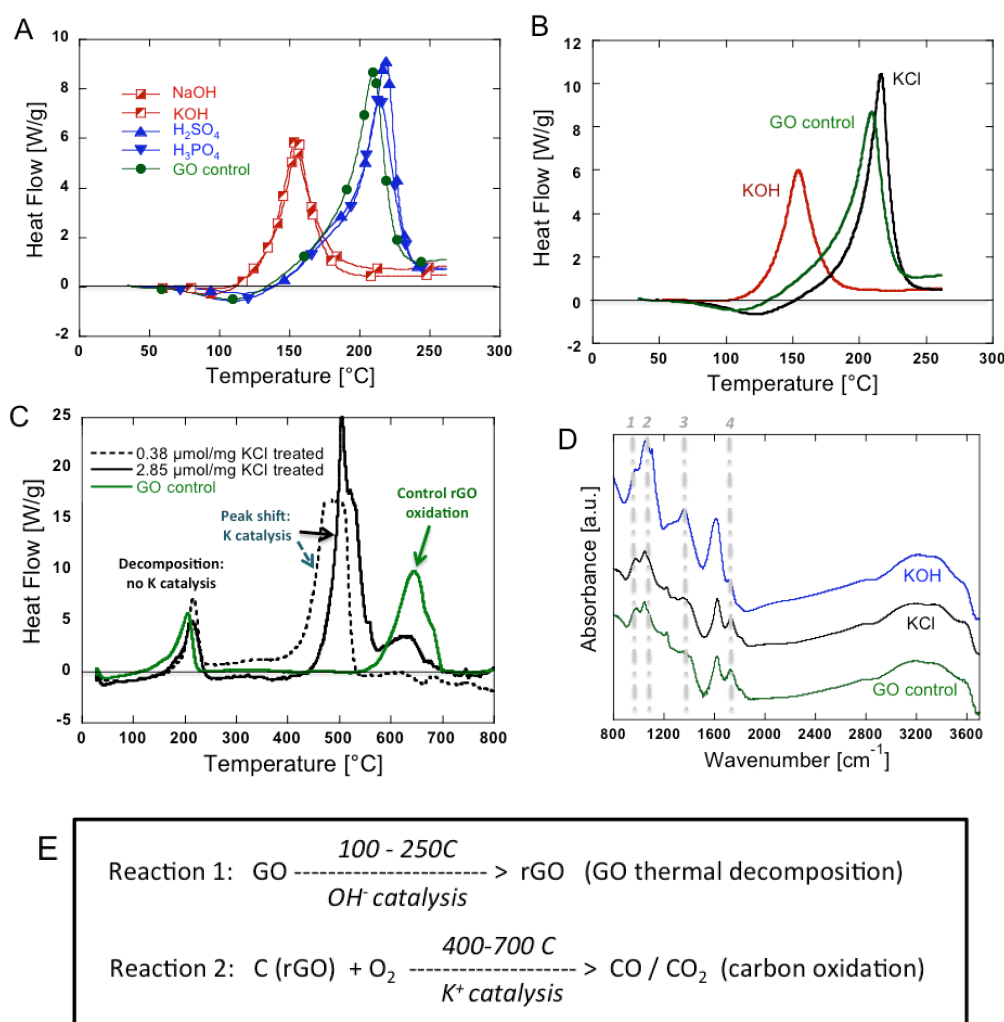


Figure 3.2 Role of salt impurities on GO decomposition and rGO combustion kinetics.

Samples were prepared by adding salts, bases, or acids into GO suspensions (GO:additive = 100:1 by mass) and dried overnight at 60°C in air. (A, B) DSC thermograms show that K^+ does not catalyze GO thermal decomposition, but OH^- does. (C) DSC thermograms on GO with salt additives in non-hermetic DSC pan in air. K^+ does not affect GO decomposition, but strongly catalyzes rGO

combustion at higher temperature. (D) FTIR spectra of GO and GO with salt additives. KCl addition has no effect on the spectrum, while KOH addition alters several peaks associated with C-O or C=O bonding. (E) Global reactions in GO thermal behavior.

The FTIR spectra in Figure 3.2D show that K^+ does not affect carbon-oxygen bonding in GO, but OH^- addition does, which may provide insight into its catalytic effects on decomposition. Specific peak interpretations for GO can be difficult due to overlapping features⁴⁰, but we can point out several specific alterations to the spectra upon OH^- addition. There is a decrease in intensity of the 1750 cm^{-1} shoulder peak (labeled “4”) relative to the adjacent 1620 cm^{-1} peak, which is commonly associated with pristine graphenic domain C=C vibrations. This same change has been seen by Taniguchi et al⁵⁰, and the reduction in the 1750 cm^{-1} peak was suggested to be related to carboxyl groups that become reversibly quenched by K^+ binding in basic solution. Dimiev et al.⁴⁰ see a similar change in this peak ratio ($1750/1620\text{ cm}^{-1}$) with addition of base and assign it to differences in water content between the two samples. Base addition to GO increases or creates a peak at $\sim 1350\text{ cm}^{-1}$ (labeled “3”), which could be C-OH⁵¹, or carboxylic acid, O=C-OH⁵². In the C-O region from $900\text{--}1200\text{ cm}^{-1}$, there is a decrease in the 970 cm^{-1} (labeled “1”) shoulder peak (possibly epoxide) relative to the main 1042 cm^{-1} (labeled “2”) peak representing various C-O structures¹⁸. These changes may represent epoxide ring-opening during chemical reduction, which is discussed in more detail later. At the same time we observed broadening of hydroxyl peak around 1040 cm^{-1} , indicating more complicated alcoholic structures form. These two changes imply the conversion from epoxides to hydroxyls during chemical reduction of GO.

Motivated by the observed effect of OH^- , we performed additional experiments to study pH effects more broadly. Figure 3.3A and B show that the pH of the original GO suspension has a strong effect on thermal decomposition kinetics. Increasing pH from 2 to 12 decreases the DSC peak onset temperature by over 50°C , which is a highly significant increase in GO

reactivity. Back-titration from pH=12 to pH=2 before drying reverses this effect – the low reactivity of the pH=2 product is restored, but only a fraction of the total exothermic heat (area under the curve) remains. This behavior suggests that OH^- catalyzes GO reduction in suspension, which has already been reported⁵³, but it further suggests that the partially reduced states captured by drying solid GO from pH=12 suspensions have an elevated thermal reactivity.

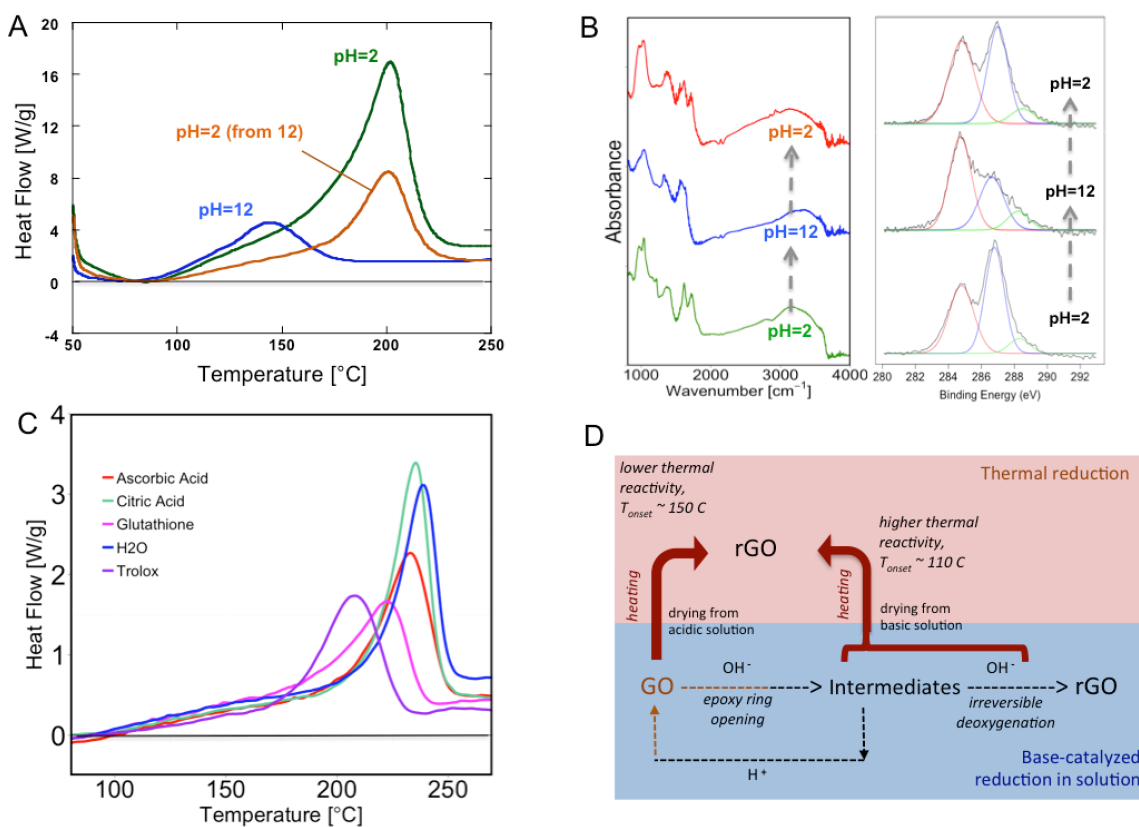


Figure 3.3 pH effects on the structural and thermal behavior of GO.

The GO samples used for these characterizations are recovered and dried from suspension stored at different pH's. (A) DSC thermograms of acidic and basic GOs. Base treatment to pH = 12 reduces GO, however lowers significantly decomposition onset temperature down to ~100°C. Same GO back treatment to pH = 2 increases enthalpy of decomposition, hence the total heat is still less than original pH = 2 GO heat of decomposition. Onset temperature moves back to ~150°C after acid treatment of GO. (B) FTIR and XPS spectra of acid and based-treated GO samples. GO base treatment to pH = 12 reduces FTIR peaks at 1050 cm⁻¹, 970 cm⁻¹, 1754 cm⁻¹ groups. GO recovered from pH = 2, back treated from pH = 12 exhibits similar but not identical functionalities as pH = 2. The de-convoluted peaks in XPS spectra are C-C/C=C (red), C-O (blue) and C=C (green), respectively. Similar to FTIR, GO back titrated from pH = 12 to pH=2 exhibits similar but not identical functionalities as initial pH = 2 sample. (C) DSC thermograms of common weak reducing agent

treated GOs. The decomposition reaction onset temperature is lowered in all four cases. (D) A semi-global kinetic structure of the coupled thermal and base-catalyzed reduction of GO: pH of precursor suspension influences reactivity and safety of thermal reduction.

The molecular mechanism is not fully understood, but FTIR and XPS provide relevant information. Figure 3.3B shows reversible changes in the C1s XPS spectra upon pH change from 2 to 12 and back-titration to 2. Similar behavior has been reported by Tanigushi et al.⁵⁰ and interpreted as base-catalyzed epoxy ring opening reactions that are reversible on addition of acid. Reversible behavior is also seen in the FTIR spectra of Figure 3.3B – the spectral changes that occur on base addition, discussed already prior (and seen in Figure 3.2D), disappear upon back titration, and the pH=2 spectrum is essentially identical before and after the titration cycle. Taniguchi et al. present a theory for the reversible behavior based on pH-dependent epoxy ring opening/closing reactions⁵⁰. Dimiev et al. also discuss base catalyzed epoxy ring opening reactions to form vicinal diols⁴⁰, which react further and irreversibly involving C-C bond cleavage and formation of vinylogous carboxylic acids. Further reaction in basic solution leads to CO₂ release, GO deoxygenation, and eventual degradation of GO into humic-like substances⁴⁰. The present paper does not shed new insight into this molecular mechanism, but does show for the first time that these chemical transformations lead to increased thermal reactivity in GO when the intermediate, partially reduced states are captured by drying. This is consistent with the proposed mechanisms of base-catalyzed GO transformation, which involve defect creation during C-C bond breakage⁴⁰. The elevated reactivity of partially reduced GO has important implications for the maximum allowed temperatures for safe processing of dry GO. A semi-global reaction scheme that is consistent with our data and the recent literature is shown in Figure 3.3D.

A series of experiments were carried out to test if only OH⁻ affects the thermal reactivity of dried GO, or if other chemical reducing agents have similar effects. Figure 3.3C shows a series of DSC traces on GO samples before and after incubation with a variety of

chemical reducing agents (glutathione, ascorbic acid, citric acid, and Trolox). In each case, partial chemical reduction lowers the enthalpy of thermal decomposition, but raises the reactivity, seen as a lowering of the thermal decomposition onset temperature. The effect of base seen in Figure 3.3C appears to be a general effect of partial chemical reduction. In addition, NaBH_4 and hydrazine have also been tested, which exhibit similar effect (Figure 3.4). The elevated reactivity is seen when the chemical reducing agent is left in the dried GO, but also when it is removed by washing, though less significantly. This suggests that the source of the elevated rGO reactivity lies in the transformed chemical structure of the partially rGO, and not the presence of the chemical agent during heating.

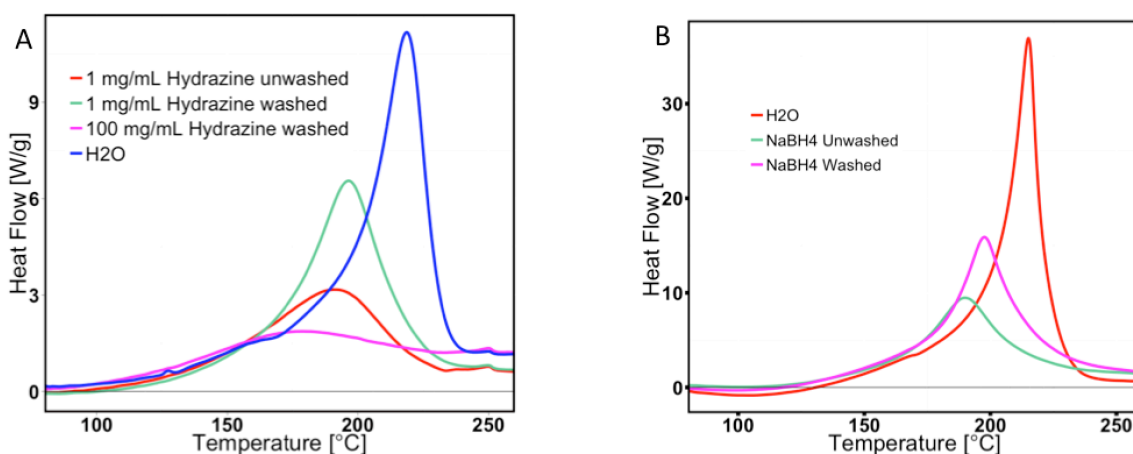


Figure 3.4 Reduction effects on the thermal behavior of GO.

The GO samples used for these characterizations are recovered and dried from suspension incubated with hydrazine or NaBH_4 . DSC thermograms of hydrazine (A) or NaBH_4 (B) treated GO. In both cases, partial reduction lowers the enthalpy of decomposition but at the same time shifts the onset temperature lower. The washed sample shows similar behavior. Complete reduction with high concentration reducing agents however removes most of the exothermicity probably due to the removal of active oxygen function groups.

3.3.3 Effect of GO aging

There have been several publications, which report that GO molecular structure evolves over the time during storage in the dry state or in suspension³⁷⁻⁴¹. One study reports that

GO aging involves loss of reactive epoxide groups³⁸. However, it seems that epoxide groups are likely source of the high formation energy for GO structure, and therefore high decomposition energy (see our group contribution calculations results in Figure 3.5A). We thus initially suspected that aging would reduce the heat and/or reactivity associated with thermal decomposition. The results in Figure 3.5B show that in approximately three-month period this is not the case. Storage of GO in ambient conditions for up to 82 days has no significant effect on the decomposition enthalpy, and the reactivity shows a slight increase (measured as slight decrease in exotherm onset temperature). Therefore, exothermic decomposition and explosion hazards are not just features of freshly prepared GO, but also of aged GO stored up to at least several months.

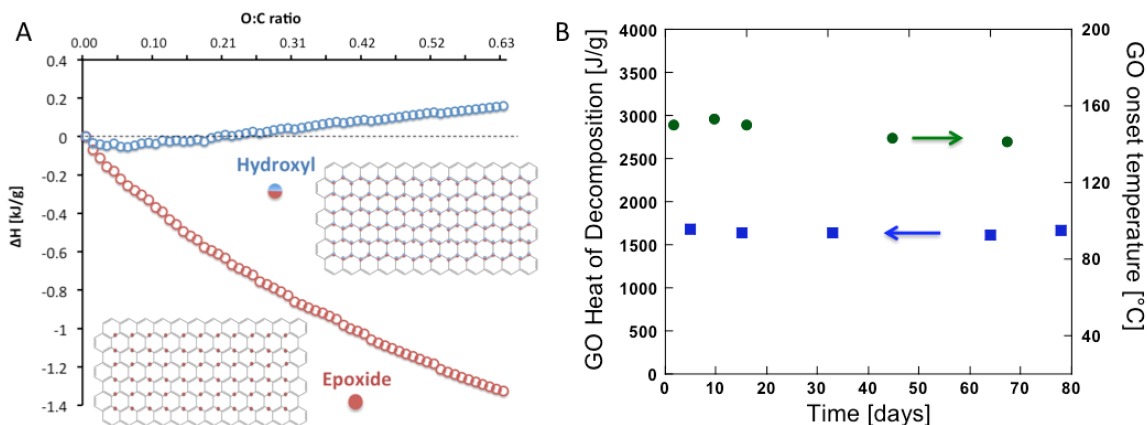


Figure 3.5 Origin of the exothermicity and aging effects of GO thermal reduction.

(A) Theoretical decomposition enthalpies of potential GO structures with varying O:C ratio, and the amount of either epoxides (structure shown in lower left inset) or hydroxyls (structure shown in upper right inset) estimated applying calculated enthalpies of formation of GO (from group contribution method). Increasing epoxide content makes GO decomposition significantly more exothermic, while increasing hydroxyl content makes GO decomposition slightly endothermic. (B) The effect of GO aging on thermal decomposition enthalpy and reactivity. In approximately three months time frame GO aging does not significantly affect the decomposition enthalpy but does slightly lower the GO onset temperature.

3.3.4 Results of the decomposition kinetics analysis

Calculation methods are needed to define safe storage and processing conditions for a variety of sample sizes, shapes, and external environments. Such calculations require quantitative decomposition kinetics data, which we derive here from DSC data taken with systematic variation of heating rate. Results in Figure 3.6A show a regular shift in DSC trace with increasing heating rate. The kinetic analysis applied here is based on peak maximum temperature⁴² recorded at each heating rate to give activation energy and frequency factor as defined in Eq. (1). The activation energy obtained in this work (142 kJ/mol) is similar to those reported previously using similar or different methods^{46, 54}. By analyzing dynamic mode DSC data the apparent reaction order n was estimated to be 0.8. The latter value was then corrected to be 0.7, applying actual isothermal mode DSC data and the kinetic parameters determined earlier (see results in Figure 3.6C).

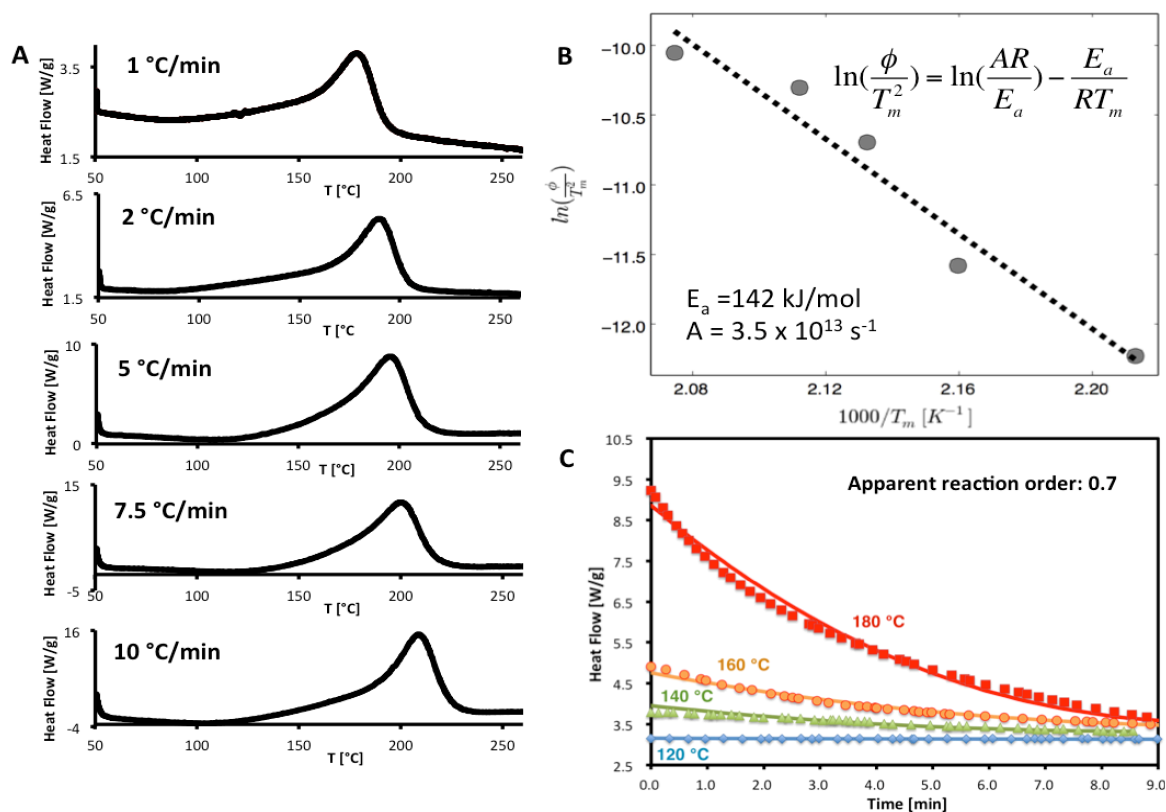


Figure 3.6 Quantitative determination of GO thermal decomposition kinetics.

The GO used here is as-prepared GO stored at room temperature in atmosphere. The experiments were carried out with small sample sizes to avoid the explosive mode of decomposition. (A) Dynamic mode DSC thermograms with systematic variation in heating rate. (B) Extraction of quantitative kinetics parameters. (C) Validation of theoretical kinetic data obtained (activation energy, pre-exponential factor and rate order). Experimental isothermal DSC data (data points) compared with the theoretical curve (solid line). The determined kinetic model matches well with experimental data.

3.3.5 Discussion of results

Thermal reduction of GO is clearly a complex process, and results in Figures 1-5 provide some insight into molecular mechanism and have direct implications for safety in storage and processing. Thermal “reduction” is in reality thermal decomposition or pyrolysis process⁵⁵⁻⁵⁷ and GO is unusual in that pyrolysis of oxygen-rich organic materials like coals and biomass is typically endothermic⁵⁸⁻⁶². The annealing of oxidized carbon surfaces leading to deoxygenation (or devolatilization) is also usually endothermic⁶³, but there are exceptions for some oxidized carbon materials, for example ozonized fullerene and KMnO_4 oxidized carbon nanotubes^{11, 64}. When compared with other common types of oxygen-containing carbon materials, the most significant structural differences of GO is the presence of large amount of epoxide⁶⁵, which are reactive groups with bond strain³⁸, and are not typically the primary functional group on carbon surfaces that are the results of reactions with ambient O_2 . Our group contribution calculations (Figure 3.5A) suggest that epoxides are the source of the high formation energy of GO and thus its exothermic decomposition. A common feature of oxidized carbons that decompose exothermically appears to be the use of certain oxygen ring-creating oxidants like ozone, which forms highly reactive ozonides, or permanganate, which is a well-known catalyst for epoxide formation in synthetic organic chemistry⁶⁶⁻⁶⁷. Ozonized fullerene and KMnO_4 oxidized

carbon nanotubes both show exothermic decomposition under heating ^{11,64}, which supports our calculation-based argument about the epoxide role in exothermic decomposition of GO.

This work further suggests that epoxides are involved in the complex response of GO to suspension pH. Base treatment renders GO more reactive to thermal decomposition, and back titration to low pH partially reverses the effect. Reversible pH effects have been seen by Taniguchi are attributed to epoxide ring opening/closing ⁵⁰, while the present paper shows for the first time that these reversible transformations also affect thermal decomposition reactivity. Drying GO from basic solutions can produce a more reactive solid product that begins to decompose at lower temperatures – in some cases down into the range of common drying operations, which can become a special safety concern. The reactive species may be vicinal diols resulting from epoxy ring opening, and these can further cleave to produce ketones or aldehydes ⁶⁸. Ketones and aldehydes are capable of further conversion to carboxyls under alkaline or thermal treatment ^{40,69}. Diols can also be converted back to cyclic ethers including epoxides in acidic environments, leading to the partial reversibility shown in Figure 3.3D. The high thermal reactivity of the diol relative to epoxy is consistent with molecular dynamics and DFT simulations of GO thermal decomposition ^{41, 69} that show lower activation barriers for the initial step of OH decomposition compared to epoxy.

The observation of elevated thermal reactivity in partially reduced GO may be more general. It occurs not only following OH⁻ treatment but also for a variety of chemical reducing agents and reduction pathways (Figure 3.3C). Ahn et al. have also observed high reactivity in partially reduced GO samples ⁷⁰.

As mentioned by previous theoretical studies ^{41,69,71}, the energy of GO is lowered by clustering of the oxidized sites into domains, which co-exist with pristine sp² domains.

Partial reduction here may break up the oxidized domains leading to more isolated oxygen-containing functional groups with high reactivity.

Of course, extensive or complete reduction eliminates the exotherm completely, as seen in results offered on Figure 3.1S (GO treatment with high concentration hydrazine). The phenomenon of elevated reactivity in GO samples after partial chemical reduction deserves further study.

Finally, the new kinetic parameters allow estimations of expected self-heating behavior relevant to industrial safety (Figure 3.7). An unsteady heat balance on a uniform GO mass gives the differential equation seen in Figure 3.7A, which can be solved numerically to give the predictions offered in Figure 3.7B,C. The simplest case is adiabatic self-heating, in which all of the reaction heat is retained in the GO body and the system always reaches the theoretical adiabatic temperature: $\Delta T = \Delta H/C_p$, which here is $> 2,000^\circ\text{C}$ for C_p around $0.5 \text{ J g}^{-1}\text{K}^{-1}$. The temperature behavior (see Figure 3.7B) shows first an incubation period of variable time length, followed by the temperature upturn, which leads to a thermal runaway. At room temperature the incubation times are very long, but at temperatures above $\sim 150^\circ\text{C}$ runaway can happen in minutes.

Adiabatic self-heating analysis is a crude estimate that only gives satisfactory result for fast processes that occur with short time for heat loss.

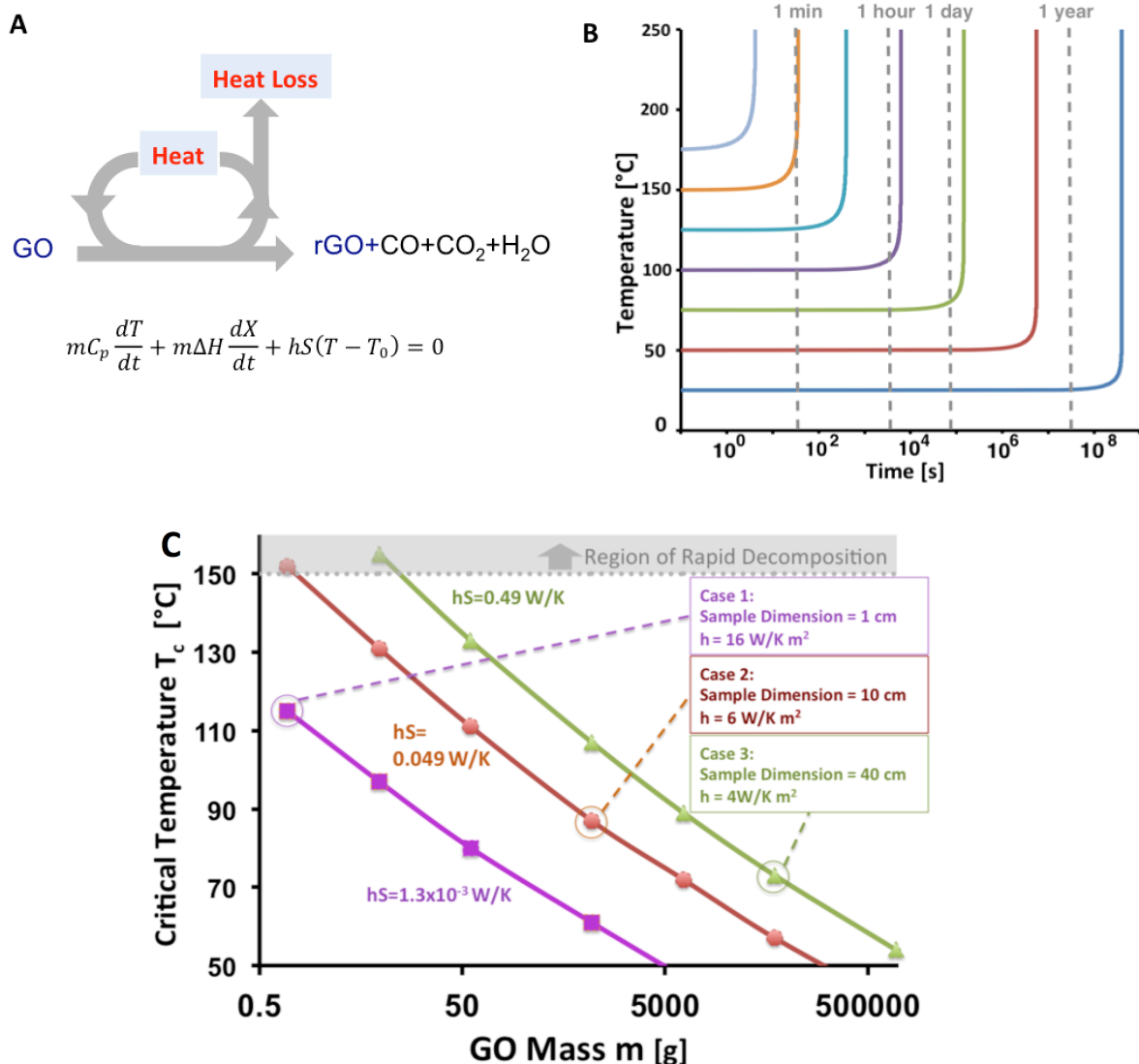


Figure 3.7 Self-heating behavior of large GO samples during storage.

(A) Differential energy balance and decomposition mechanism (B) Temperature curves for numerical solutions for adiabatic self-heating case, (C) T_c vs m for non-adiabatic self-heating cases as a function of heat transfer coefficient and surface area product, hS ; T_c is the critical initial temperature above which the runaway occurs and below which a steady-state temperature is achieved; m , the mass of GO; h , the heat transfer coefficient; S , the external surface area. GO with greater mass and smaller value of hS is more likely to reach the point of no return to initiate the thermal runaway reaction. Three example cases are shown with different sample dimensions and associated typical heat transfer coefficients when heat convection from the GO surface to the environment is the rate limiting in heat transfer mechanism.

The results of non-adiabatic analysis are more practical. The method applies specific values of heat transfer coefficient (h) and the GO external surface area (S). In some cases this analysis gives a steady state temperature, while in other cases it gives an incubation

period followed by an upturn and thermal runaway. Figure 3.7C plots the critical temperature T_c above, which the runaway occurs (and below which a steady-state temperature is achieved and no runaway) as a function of the initial bulk GO mass m . Some typical values for large and small GO forms (sample dimension in centimetres) and typical heat transfer coefficients (assuming GO to be a sphere) affecting critical temperature are shown in Figure 3.7C. As seen from this figure larger in size GO pieces have lower critical temperatures.

3.4 Conclusions

The energetic nature of GO was first identified long ago, but a significant body of new research is needed to understand it at a level that will allow the safe management of this behavior in large-scale nano-manufacturing of graphene-based materials. This article presents an experimental characterization of the thermochemistry of GO, and reports quantitative values for the enthalpy of combustion, the enthalpy of thermal decomposition, the rates of thermal decomposition, and their dependence on temperature, GO composition, and processing history. GO decomposition is shown to be accelerated by partial chemical reduction, or by exposure to high-pH conditions in suspension prior to drying. Transformations of epoxide groups are suggested as the cause of the exothermic decomposition and the reversible effects of acid/base treatment. Residual potassium does not catalyze GO decomposition, but rather catalyzes the subsequent heterogeneous combustion of the rGO carbon product. This body of data suggests that large samples of GO in some storage and handling scenarios will be capable of self-heating and spontaneous decomposition leading to high-volume gas release and explosive events. The quantitative kinetic parameters reported here can be used to identify safe conditions for storage and handling, and to identify compositions and processing protocols that will help ensure the

successful transition of GO-based graphene materials from laboratory subjects to commodity products.

3.5 References

1. Brodie, B. C., On the atomic weight of graphite. *Philos. Trans. R. Soc. London* 1859, 249-259.
2. Scholz, W.; Boehm, H. P., Die Thermische Zersetzung Von Graphitoxyd. *Naturwissenschaften* 1964, 51 (7), 160.
3. Boehm, H. P.; Scholz, W., Der Verpuffungspunkt Des Graphitoxids. *Z. Anorg. Allg. Chem.* 1965, 335 (1-2), 74.
4. Boehm, H.; Clauss, A.; Fischer, G.; Hofmann, U. In *Surface properties of extremely thin graphite lamellae*, Proceedings of the fifth Conference on Carbon, Pergamon Press: 1962; pp 73-80.
5. Qiu, Y.; Guo, F.; Hurt, R.; Külaots, I., Explosive thermal reduction of graphene oxide-based materials: Mechanism and safety implications. *Carbon* 2014, 72 (0), 215–223.
6. Kim, J.; Cote, L. J.; Kim, F.; Huang, J., Visualizing graphene based sheets by fluorescence quenching microscopy. *J. Am. Chem. Soc.* 2010, 132 (1), 260-7.
7. Krishnan, D.; Kim, F.; Luo, J. Y.; Cruz-Silva, R.; Cote, L. J.; Jang, H. D.; Huang, J. X., Energetic graphene oxide: Challenges and opportunities. *Nano Today* 2012, 7 (2), 137-152.
8. Botas, C.; Alvarez, P.; Blanco, C.; Santamaria, R.; Granda, M.; Gutierrez, M. D.; Rodriguez-Reinoso, F.; Menendez, R., Critical temperatures in the synthesis of graphene-like materials by thermal exfoliation-reduction of graphite oxide. *Carbon* 2013, 52, 476-485.

9. You, S. J.; Luzan, S. M.; Szabo, T.; Talyzin, A. V., Effect of synthesis method on solvation and exfoliation of graphite oxide. *Carbon* 2013, 52, 171-180.
10. McAllister, M. J.; Li, J. L.; Adamson, D. H.; Schniepp, H. C.; Abdala, A. A.; Liu, J.; Herrera-Alonso, M.; Milius, D. L.; Car, R.; Prud'homme, R. K.; Aksay, I. A., Single sheet functionalized graphene by oxidation and thermal expansion of graphite. *Chem. Mater.* 2007, 19 (18), 4396-4404.
11. Cataldo, F.; Compagnini, G.; D'Urso, L.; Palleschi, G.; Valentini, F.; Angelini, G.; Braun, T., Characterization of Graphene Nanoribbons from the Unzipping of MWCNTs. *Fuller Nanotub Car N* 2010, 18 (3), 261-272.
12. Jimenez, P. S. V., Thermal-Decomposition of Graphite Oxidation-Products Dsc Studies of Internal-Combustion of Graphite Oxide. *Mater. Res. Bull.* 1987, 22 (5), 601-608.
13. Novoselov, K. S.; Jiang, D.; Schedin, F.; Booth, T. J.; Khotkevich, V. V.; Morozov, S. V.; Geim, A. K., Two-dimensional atomic crystals. *Proc. Natl. Acad. Sci. U. S. A.* 2005, 102 (30), 10451-10453.
14. Novoselov, K. S.; Geim, A. K.; Morozov, S. V.; Jiang, D.; Zhang, Y.; Dubonos, S. V.; Grigorieva, I. V.; Firsov, A. A., Electric field effect in atomically thin carbon films. *Science* 2004, 306 (5696), 666-669.
15. Geim, A. K.; Novoselov, K. S., The rise of graphene. *Nat Mater* 2007, 6 (3), 183-191.
16. Chen, C. M.; Yang, Q. H.; Yang, Y. G.; Lv, W.; Wen, Y. F.; Hou, P. X.; Wang, M. Z.; Cheng, H. M., Self-Assembled Free-Standing Graphite Oxide Membrane. *Adv. Mater.* 2009, 21 (29), 3007-+.
17. Kim, J.; Cote, L. J.; Kim, F.; Yuan, W.; Shull, K. R.; Huang, J. X., Graphene Oxide Sheets at Interfaces. *J. Am. Chem. Soc.* 2010, 132 (23), 8180-8186.
18. Xu, Y. X.; Sheng, K. X.; Li, C.; Shi, G. Q., Self-Assembled Graphene Hydrogel via a One-Step Hydrothermal Process. *ACS nano* 2010, 4 (7), 4324-4330.

19. Huang, H.; Chen, P. W.; Zhang, X. T.; Lu, Y.; Zhan, W. C., Edge-to-Edge Assembled Graphene Oxide Aerogels with Outstanding Mechanical Performance and Superhigh Chemical Activity. *Small* 2013, 9 (8), 1397-1404.
20. Zhao, G. X.; Li, J. X.; Ren, X. M.; Chen, C. L.; Wang, X. K., Few-Layered Graphene Oxide Nanosheets As Superior Sorbents for Heavy Metal Ion Pollution Management. *Environ. Sci. Technol.* 2011, 45 (24), 10454-10462.
21. Stankovich, S.; Dikin, D. A.; Dommett, G. H. B.; Kohlhaas, K. M.; Zimney, E. J.; Stach, E. A.; Piner, R. D.; Nguyen, S. T.; Ruoff, R. S., Graphene-based composite materials. *Nature* 2006, 442 (7100), 282-286.
22. Singh, V. K.; Shukla, A.; Patra, M. K.; Saini, L.; Jani, R. K.; Vadera, S. R.; Kumar, N., Microwave absorbing properties of a thermally reduced graphene oxide/nitrile butadiene rubber composite. *Carbon* 2012, 50 (6), 2202-2208.
23. Zhu, P. S.; Huang, S. S.; Li, M. Y.; Ding, N.; Peng, B.; Kong, L. M.; Bo, Y., A Sandwiched Biological Fluorescent Probe for the Diagnosis of Human Ovarian Tumor Based on TiO₂ Nanoparticles. *Journal of Fluorescence* 2011, 21 (1), 179-186.
24. Zhang, W.; Gong, X.; Cai, Y.; Zhang, C.; Yu, X.; Fan, J.; Diao, G., Investigation of water-soluble inclusion complex of hypericin with beta-cyclodextrin polymer. *Carbohydr. Polym.* 2013, 95 (1), 366-70.
25. Chen, G. H.; Weng, W. G.; Wu, D. J.; Wu, C. L., PMMA/graphite nanosheets composite and its conducting properties. *Eur. Polym. J.* 2003, 39 (12), 2329-2335.
26. Gokturk, H. S.; Fiske, T. J.; Kalyon, D. M., Effects of Particle-Shape and Size Distributions on the Electrical and Magnetic-Properties of Nickel/Polyethylene Composites. *J. Appl. Polym. Sci.* 1993, 50 (11), 1891-1901.
27. Fiori, G.; Bonaccorso, F.; Iannaccone, G.; Palacios, T.; Neumaier, D.; Seabaugh, A.; Banerjee, S. K.; Colombo, L., Electronics based on two-dimensional materials. *Nat Nanotechnol* 2014, 9 (10), 768-779.

28. Liu, J., Charging graphene for energy. *Nat Nanotechnol* 2014, 9 (10), 739-741.
29. Torrisi, F.; Coleman, J. N., Electrifying inks with 2D materials. *Nat Nanotechnol* 2014, 9 (10), 738-739.
30. Ahn, J. H.; Hong, B. H., Graphene for displays that bend. *Nat Nanotechnol* 2014, 9 (10), 737-738.
31. Bohm, S., Graphene against corrosion. *Nat Nanotechnol* 2014, 9 (10), 741-742.
32. Kim, F.; Luo, J. Y.; Cruz-Silva, R.; Cote, L. J.; Sohn, K.; Huang, J. X., Self-Propagating Domino-like Reactions in Oxidized Graphite. *Adv. Funct. Mater.* 2010, 20 (17), 2867-2873.
33. Zhang, X. Y.; Huang, Y.; Wang, Y.; Ma, Y. F.; Liu, Z. F.; Chen, Y. S., Synthesis and characterization of a graphene-C-60 hybrid material. *Carbon* 2009, 47 (1), 334-337.
34. Becerril, H. A.; Mao, J.; Liu, Z.; Stoltenberg, R. M.; Bao, Z.; Chen, Y., Evaluation of solution-processed reduced graphene oxide films as transparent conductors. *ACS nano* 2008, 2 (3), 463-470.
35. Senthilkumaar, S.; Varadarajan, P. R.; Porkodi, K.; Subbhuraam, C. V., Adsorption of methylene blue onto jute fiber carbon: kinetics and equilibrium studies. *J. Colloid Interface Sci.* 2005, 284 (1), 78-82.
36. Watcharotone, S.; Dikin, D. A.; Stankovich, S.; Piner, R.; Jung, I.; Dommett, G. H. B.; Evmenenko, G.; Wu, S. E.; Chen, S. F.; Liu, C. P.; Nguyen, S. T.; Ruoff, R. S., Graphene-silica composite thin films as transparent conductors. *Nano Lett.* 2007, 7 (7), 1888-1892.
37. Chua, C. K.; Pumera, M., Light and Atmosphere Affect the Quasi-equilibrium States of Graphite Oxide and Graphene Oxide Powders. *Small* 2014.
38. Kim, S.; Zhou, S.; Hu, Y. K.; Acik, M.; Chabal, Y. J.; Berger, C.; de Heer, W.; Bongiorno, A.; Riedo, E., Room-temperature metastability of multilayer graphene oxide films. *Nat Mater* 2012, 11 (6), 544-549.

39. Pendolino, F.; Parisini, E.; Lo Russo, S., Time-Dependent Structure and Solubilization Kinetics of Graphene Oxide in Methanol and Water Dispersions. *The Journal of Physical Chemistry C* 2014.
40. Dimiev, A. M.; Alemany, L. B.; Tour, J. M., Graphene oxide. Origin of acidity, its instability in water, and a new dynamic structural model. *ACS nano* 2013, 7 (1), 576-88.
41. Zhou, S.; Bongiorno, A., Origin of the chemical and kinetic stability of graphene oxide. *Sci. Rep.* 2013, 3, 2484.
42. Kissinger, H. E., Reaction Kinetics in Differential Thermal Analysis. *Anal. Chem.* 1957, 29 (11), 1702-1706.
43. Marrero, J.; Gani, R., Group-contribution based estimation of pure component properties. *Fluid Phase Equilib.* 2001, 183, 183-208.
44. Henrich, E.; Burkle, S.; Meza-Renken, Z. I.; Rumpel, S., Combustion and gasification kinetics of pyrolysis chars from waste and biomass. *J. Anal. Appl. Pyrolysis* 1999, 49 (1-2), 221-241.
45. Stanmore, B. R.; Brilhac, J. F.; Gilot, P., The oxidation of soot: a review of experiments, mechanisms and models. *Carbon* 2001, 39 (15), 2247-2268.
46. Jung, I.; Field, D. A.; Clark, N. J.; Zhu, Y. W.; Yang, D. X.; Piner, R. D.; Stankovich, S.; Dikin, D. A.; Geisler, H.; Ventrice, C. A.; Ruoff, R. S., Reduction Kinetics of Graphene Oxide Determined by Electrical Transport Measurements and Temperature Programmed Desorption. *J Phys Chem C* 2009, 113 (43), 18480-18486.
47. Antal, M. J.; Gronli, M., The art, science, and technology of charcoal production. *Ind. Eng. Chem. Res.* 2003, 42 (8), 1619-1640.
48. Jones, J. M.; Darvell, L. I.; Bridgeman, T. G.; Pourkashanian, M.; Williams, A., An investigation of the thermal and catalytic behaviour of potassium in biomass combustion. *P Combust Inst* 2007, 31, 1955-1963.

49. Jensen, A.; Dam-Johansen, K.; Wojtowicz, M. A.; Serio, M. A., TG-FTIR study of the influence of potassium chloride on wheat straw pyrolysis. *Energy Fuels* 1998, *12* (5), 929-938.
50. Taniguchi, T.; Kurihara, S.; Tateishi, H.; Hatakeyama, K.; Koinuma, M.; Yokoi, H.; Hara, M.; Ishikawa, H.; Matsumoto, Y., pH-driven, Reversible Epoxy Ring Opening/Closing in Graphene Oxide. *Carbon* 2015, *84*, 560–566.
51. Yan, J.; Wei, T.; Shao, B.; Fan, Z. J.; Qian, W. Z.; Zhang, M. L.; Wei, F., Preparation of a graphene nanosheet/polyaniline composite with high specific capacitance. *Carbon* 2010, *48* (2), 487-493.
52. Acik, M.; Lee, G.; Mattevi, C.; Pirkle, A.; Wallace, R. M.; Chhowalla, M.; Cho, K.; Chabal, Y., The Role of Oxygen during Thermal Reduction of Graphene Oxide Studied by Infrared Absorption Spectroscopy. *The Journal of Physical Chemistry C* 2011, *115* (40), 19761-19781.
53. Fan, X. B.; Peng, W. C.; Li, Y.; Li, X. Y.; Wang, S. L.; Zhang, G. L.; Zhang, F. B., Deoxygenation of Exfoliated Graphite Oxide under Alkaline Conditions: A Green Route to Graphene Preparation. *Adv. Mater.* 2008, *20* (23), 4490-4493.
54. Kuibo, Y.; Haitao, L.; Yidong, X.; Hengchang, B.; Jun, S.; Zhiguo, L.; Litao, S., Thermodynamic and kinetic analysis of low-temperature thermal reduction of graphene oxide. *Nano-Micro Letters* 2011, *3* (1), 51-55.
55. Lin, Z. Y.; Waller, G.; Liu, Y.; Liu, M. L.; Wong, C. P., Facile Synthesis of Nitrogen-Doped Graphene via Pyrolysis of Graphene Oxide and Urea, and its Electrocatalytic Activity toward the Oxygen-Reduction Reaction. *Adv Energy Mater* 2012, *2* (7), 884-888.
56. Stankovich, S.; Dikin, D. A.; Piner, R. D.; Kohlhaas, K. A.; Kleinhammes, A.; Jia, Y.; Wu, Y.; Nguyen, S. T.; Ruoff, R. S., Synthesis of graphene-based nanosheets via chemical reduction of exfoliated graphite oxide. *Carbon* 2007, *45* (7), 1558-1565.

57. Compton, O. C.; Nguyen, S. T., Graphene Oxide, Highly Reduced Graphene Oxide, and Graphene: Versatile Building Blocks for Carbon-Based Materials. *Small* 2010, 6 (6), 711-723.
58. Mahajan, O. P.; Tomita, A.; Walker, P. L., Differential Scanning Calorimetry Studies on Coal .1. Pyrolysis in an Inert Atmosphere. *Fuel* 1976, 55 (1), 63-69.
59. Mohan, D.; Pittman, C. U.; Steele, P. H., Pyrolysis of wood/biomass for bio-oil: A critical review. *Energy Fuels* 2006, 20 (3), 848-889.
60. Antal, M. J.; Varhegyi, G., Cellulose Pyrolysis Kinetics - the Current State Knowledge. *Ind. Eng. Chem. Res.* 1995, 34 (3), 703-717.
61. Bradbury, A. G. W.; Sakai, Y.; Shafizadeh, F., A kinetic model for pyrolysis of cellulose. *J. Appl. Polym. Sci.* 1979, 23 (11), 3271-3280.
62. Kok, M. V.; Ozbas, E.; Karacan, O.; Hicyilmaz, C., Effect of particle size on coal pyrolysis. *J. Anal. Appl. Pyrolysis* 1998, 45 (2), 103-110.
63. Milosavljevic, I.; Oja, V.; Suuberg, E. M., Thermal effects in cellulose pyrolysis: Relationship to char formation processes. *Ind. Eng. Chem. Res.* 1996, 35 (3), 653-662.
64. Heymann, D.; Bachilo, S. M.; Weisman, R. B.; Cataldo, F.; Fokkens, R. H.; Nibbering, N. M. M.; Vis, R. D.; Chibante, L. P. F., C₆₀O₃, a fullerene ozonide: Synthesis and dissociation to C₆₀O and O₂. *J. Am. Chem. Soc.* 2000, 122 (46), 11473-11479.
65. Lerf, A.; He, H. Y.; Forster, M.; Klinowski, J., Structure of graphite oxide revisited. *J. Phys. Chem. B* 1998, 102 (23), 4477-4482.
66. Hanson, J. R.; Hitchcock, P. B.; Liman, M. D.; Nagaratnam, S.; Manickavasagar, R., The Stereochemistry of Epoxidation of Some Steroidal Alkenes with Potassium Permanganate-Copper Sulfate. *J Chem Res-S* 1995, (6), 220-221.
67. Hanson, J. R.; Terry, N.; Uyanik, C., The stereochemistry of epoxidation of Delta(5)-steroids with sodium perborate and potassium permanganate. *J Chem Res-S* 1998, (1), 50-51.

68. Nicoli, B. H.; Shinn, L. A., The action of periodic acid on alpha-amino alcohols. *J. Am. Chem. Soc.* 1939, *61*, 1615-1615.
69. Bagri, A.; Mattevi, C.; Acik, M.; Chabal, Y. J.; Chhowalla, M.; Shenoy, V. B., Structural evolution during the reduction of chemically derived graphene oxide. *Nat. Chem.* 2010, *2* (7), 581-587.
70. Ahn, S. I.; Kim, K.; Kim, J. Y.; Kim, E. S.; Han, J. Y.; Eom, J. W.; Choi, K.-K.; Park, J.-C.; Kim, S.-H.; Jeong, E. D.; Choi, K. C., Reduction intermediates of graphene oxide for low temperature reduction electrode material. *Rsc Adv* 2014, *4* (43), 22476.
71. Larciprete, R.; Fabris, S.; Sun, T.; Lacovig, P.; Baraldi, A.; Lizzit, S., Dual path mechanism in the thermal reduction of graphene oxide. *J. Am. Chem. Soc.* 2011, *133* (43), 17315-21.

**CHAPTER 4 INTRINSIC CHEMICAL
PROPERTIES OF GO-BASED
MATERIALS AND THEIR POTENTIAL
AS EFFICIENT ANTIOXIDANT**

This chapter is based on the published work by Y. Qiu, Z. Wang, A. C. E. Owens, I. Kulaots, Y. Chen, A. B. Kane and R. H. Hurt, *Nanoscale*, 2014

4.1 Introduction

The planer sp^2 structure has been known ideal for stabilizing free radicals.¹ Idea graphene exclusively consists of sp^2 structure, but its perfect lattice also implies high energy barrier to break the symmetry and to introduce unpaired electrons or small molecular moieties. GO based materials usually contain a large portion of heteroatoms and defect sites though the sp^2 domain still exists as the major species. This property might make them highly efficient free radical scavenger and ideal antioxidant against reactive oxygen species (ROS), but to data there is no report on the intrinsic antioxidant property of GO based materials.

ROS play important roles in food spoilage,² oil rancidification,² polymer degradation,³ and damage to biological structures that include cell membranes, protein structures, and DNA.⁴⁻⁶ Oxidation protection is a broad area of chemical and biochemical technology with applications in nutrition,⁷ food and pharmaceutical formulations,^{5, 8-9} biomedical implants,¹⁰ topical protection,¹¹ cosmetics,⁸ metal corrosion,¹² wound healing therapies,¹³ and the long-term stabilization of chemical products and polymer materials.³ GO-based materials have been explored as two-dimensional physical barriers that restrict access of oxidants to surfaces, but the potential for graphene-based materials as *chemical agents* in oxidation protection has not been systematically explored to our knowledge.

Low temperature oxidation protection often involves destroying or inhibiting the formation of ROS, which form by the uncontrolled and undesired partial reduction of molecular oxygen in the presence of Fenton-active metal catalysts, material surfaces, or UV radiation. Carbon nanomaterials can generate ROS through surface reactions, often

involving defect or heteroatom sites,¹⁴⁻¹⁵ or through transition metal impurities.¹⁶⁻¹⁸ A variety of heteroatoms can be present in chemically modified nano carbons and these may govern ROS generation or toxicity.¹⁹⁻²¹ Carbon nanomaterials may also scavenge ROS, as reported by several studies focused on fullerenes, carbon nanotubes (CNTs) and functionalized carbon dots.²²⁻²⁸ Oxidation protection by carbon materials may involve radical adduct formation at sp² carbon sites, which delocalizes spin across the conjugated graphenic backbone²⁹ and leads to destruction of the radical following second adduct formation;^{26, 28} through electron transfer;³⁰⁻³¹ through hydrogen donation from functional groups;³² or through chelation of transitional metal ions and inhibition of Fenton-based radical generation.³³⁻³⁵ Graphene-based materials may show one or more of these important behaviors, but this has not been confirmed, and the relative activity of different members of the graphene family, or toward different radicals or ROS is unknown.

Here we hypothesize GO based materials may be antioxidant. To test this hypothesis we investigated the basic chemical activity of several representative GO based materials, namely graphene oxide (GO), reduced graphene oxide (rGO), and few-layer graphene (FLG) against various ROS: hydroxyl radical, superoxide, peroxide/peroxyl, and the model stable radicals 2,2-diphenyl-1-picrylhydrazyl (DPPH) and 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulphonic acid) (ABTS) radical cation. Hydroxyl radicals ($\cdot\text{OH}$) are generated *in situ* through Fenton chemistry or UV irradiation, and detected by electron paramagnetic resonance (EPR). Additional experiments are designed to test whether suspended graphene sheets can protect organic dyes from destruction by *in situ* generated $\cdot\text{OH}$. Multiple experiments here show that GO is an effective scavenger of $\cdot\text{OH}$ and can protect water soluble dyes and spin traps from oxidation. FLG is more active despite its lower surface area, suggesting that the main scavenging sites are on the pristine graphenic basal plane and are not associated with H-donation from hydroxyl groups. In experiments involving

UV-induced $\cdot\text{OH}$ generation, some of the protective effect is attributed to UV absorption by graphene in addition to radical scavenging. These behaviors are discussed in relations to the physical and chemical structures of GO and FLG. Finally, we demonstrate one application, in which graphene nanosack encapsulation is used to reduce ROS generation and oxidative damage that are undesired side effects when TiO_2 nanoparticles are used for UV protection.

4.2 Materials and methods

4.2.1 Materials

GO based materials were produced with the methods as described later in the method section. Few layer graphene (3-5 layers with nominal lateral dimension of 800 nm) was obtained commercially and characterized.

SP-1 graphite powder was purchased from Bay Carbon (Bay City, MI). $\text{K}_2\text{S}_2\text{O}_8$, KMnO_4 , ascorbic acid, titanium (IV) oxide (TiO_2) nanopowders (<25 nm particle size, >99.5 % trace metal basis), iron (II) sulfate heptahydrate ($\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$), reduced GSH, D-mannitol, pC60, xanthine, xanthine oxidase from bovine milk grade IV, 2,2'-azobis(2-amidinopropane) hydrochloride (AAPH), thiobarbituric acid (TBA), trichloroacetic acid (TCA), DPPH, 2-deoxy-D-ribose (deoxyribose), sodium dodecyl sulfate (SDS), ABTS, β -Nicotinamide adenine dinucleotide 2'-phosphate reduced tetrasodium salt hydrate (NADH), phenazinemethosulfate (PMS), xanthine, xanthine oxidase from bovine milk, and NBT were purchased from Sigma Chemicals Co (St. Louis, MO). Trolox (6-Hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid), hydrogen peroxide (H_2O_2), phosphate buffered saline (PBS) (pH 7.4), and EDTA were purchased from Fisher Scientific Inc. (Pittsburgh, PA). 5,5-dimethyl-1-pyrroline-N-oxide (DMPO) was purchased from

Dojindo Molecular Technologies, Inc. (Rockville, MD). ThioGlo-1 fluorescent thiol reagent was purchased from EMD Millipore Chemical (Darmstadt, Germany). Nanopure water was used throughout.

4.2.2 Characterization

The morphologies of all the materials used in this study were characterized with a LEO 1530 field-emission scanning electron microscope (SEM) and an FEI CM20 TEM. The graphenic structure was characterized with a WITEC Alpha 300S Scanning Near-field Optical Microscope with micro Raman system using the 532 nm laser. The functional groups on the graphenic plane and edges were identified using a JASCO FT/IR-4100 Fourier Transform Infrared Spectrometer. N₂ vapor isotherms at 77 K and CO₂ vapor isotherms at 273K were measured using Autosorb-1 instrument from Quantachrome Corporation from which surface areas were calculated by applying Brunauer Emmett Teller (BET) model, and pore size distributions by using carbon slit pore model nonlocal density functional theory (NL-DFT).³⁶ The size of TiO₂ nanoparticles were found to be around 20~30 nm with TEM and about 57 nm with dynamic light scattering (DLS). The dominant phase was identified to be anatase with X-ray Diffraction (XRD) pattern on a Bruker AXS D8 Advance instrument with Cu KR radiation ($\lambda = 1.5418 \text{ \AA}$) (see SI).

4.2.3 Methods

4.2.3.1 GO based materials preparation

GO was synthesized using a modified Hummers method as described in the last chapter. The rGO used in this study was prepared through thermal reduction, i.e., rapid heating GO

cake to 250 °C in nitrogen flow and keeping that temperature for 30 min. An explosive exfoliation was observed during its preparation.

4.2.3.2 Dye protection assay for •OH scavenging

Phenol red was used as a model target molecule for oxidant attack, and the effects of graphene-based materials on its decoloration rate in the presence of hydroxyl radical used as a measure of antioxidant activity. 10 mL solutions (pH 6.2) containing phenol red (0.1 mM), FeCl₂ (0.5 μM), H₂O₂ (50 mM) and graphene-based materials (0.10 g L⁻¹) in PBS (50 mM) were continuously stirred on a rotator for 8 hours. 1.5 mL solutions were taken out, centrifuged at 12,000 rpm to remove graphene-based materials and then the absorption spectra of the supernatant were measured with a JASCO V-630 UV/Vis Spectrometer.

4.2.3.3 •OH scavenging assay by EPR

EPR spectra were obtained using a Bruker EMXplus spectrometer with procedures similar to our previous research.³⁷ Spectra were obtained at center field of 3519 G; sweep width 100 G; microwave power 2 mW; sweep time 30 s; number of scans 10; modulation amplitude 1 G; time constant, 81.92 ms. •OH were generated *in situ* through three methods: Fenton reactions, UV photolysis and sonochemical cleavage of H₂O₂. In the former method, 180 μL solution containing DMPO (11 mM), H₂O₂ (1.1 mM), graphene-based materials (GO, rGO, FLG, etc) (up to 222 ppm) in PBS (5.5 mM, pH 3) was prepared in a centrifuge tube. Fe(II) (20 μL, 1.0 ppm) aqueous solution freshly prepared from FeSO₄ was then added to initiate the reaction. After brief stirring, about 50 μL of solution was then transferred into a capillary tube, which was in turn mounted onto the EPR spectrometer for scanning. The scanning started exactly 20 min after the addition of Fe (II) solution. In the latter method, a solution containing DMPO (1 mM), H₂O₂ (1 mM), GO aqueous solution (up to 20 ppm) in PBS

(100 mM, pH 7.4) in a quartz cuvette was exposed to UV light (365 nm at $\sim 10 \text{ mW cm}^{-2}$) for 20 min. 50 μL of the solution was then transferred into a capillary tube, which was in turn mounted onto the EPR spectrometer for scanning.

4.2.3.4 Non-enzymatic superoxide radical scavenging assay

The interaction of graphene-based materials with superoxide anion was investigated using the NADH/PMS reaction system to generate superoxide in a method similar to Valentao et al.³⁸ In a 96-well plate, 200 μL solution containing NADH (200 μM), NBT (50 μM), and test materials (100 ppm) in PBS (20 mM) was prepared and the reaction initiated by addition of PMS (40 μL , 15 μM). The absorption at 560 nm was monitored with a Molecular Device SpectraMax M2 Multimode Microplate Reader for 10 min at room temperature.

4.2.3.5 Enzymatic superoxide radical scavenging assay

Here superoxide radicals were generated using the xanthine/xanthine oxidase system using a procedure similar to Valentao et al.³⁸ In 96-well plates, 200 μL solution containing xanthine (2.0 mM), NBT (50 μM), sample solution (100 ppm), EDTA (0.5 mM) in PBS (20 mM) was prepared and the reaction initiated by addition of xanthine oxidase (5 μL , 20 unit mL^{-1}). Optical absorption at 560 nm was monitored with Molecular Device SpectraMax M2 Multimode Microplate Reader for 10 min at room temperature.

4.2.3.6 DPPH• and ABTS•⁺ scavenging assay

DPPH• and ABTS•⁺ are both relatively stable radicals and are often used to assess antioxidant activity of substances.³⁹⁻⁴⁰ A procedure similar to that reported by Fukumoto et al.³⁴ was used here in which DPPH• concentration is monitored through its characteristic absorption around 515 nm. 1000 μL of DPPH• (200 μM) solution freshly

prepared with 80% methanol and PBS (20 mM, pH 7.4) were mixed with 111 μL aqueous suspensions of the test samples (1.0 g L^{-1}). The mixtures were then left in the dark at room temperature (25°C) and the optical absorption at 516 nm were measured at regular time intervals up to 680 min.

A modified procedure from Re et al ⁴¹ was applied to serve similar purpose with $\text{ABTS}^{\bullet+}$ here. Briefly, ABTS (7 mM) aqueous solution was prepared and activated by potassium persulfate at room temperature for 12-16 hours before use. The activated $\text{ABTS}^{\bullet+}$ radical is stable in the dark for up to 2 days. The free radical solution is diluted with PBS (100 mM, pH 7.4) to get a solution with absorbance of around 0.70 at 734 nm. The aqueous solution of tested materials (100 μL , 2000 ppm) or nanopure water (100 μL) as control was added into the above solution (1.0 mL). The absorbance is recorded every 1 min after the addition of the tested solution.

4.2.3.7 GSH competitive oxidation assay

Experiments were performed to test whether the presence of graphene-based materials in suspension could protect GSH from oxidation by hydrogen peroxide. The extent of GSH oxidation was determined by measuring unreacted thiol groups using ThioGlo 1 fluorescent reagent.¹⁴ Briefly, a solution containing GSH (4 mM), H_2O_2 (1 mM), and graphene-based materials (40 ppm) in PBS (100 mM, pH 7.4) were stirred with rotator for 2 h and then filtered through Amicon Ultra-4 Centrifugal Filter Unit with an Ultracel-3 membrane. The filtrates were collected and incubated with ThioGlo-1 (20 μM) in the dark for 30 min after which fluorescence emission was measured at 513 nm (excitation at 379 nm) with the SpectraMax M2 Microplate Reader.

4.2.3.8 MDA assay

The effect of graphene-based materials on the $\cdot\text{OH}$ -mediated oxidation of deoxyribose was studied using an MDA assay modified from Halliwell and Gutteridge.⁴² Briefly, freshly prepared deoxyribose (100 mM) solution was mixed with Fenton reagents (FeCl_2 and H_2O_2) and FLG or GO suspension to get a solution containing FLG or GO (100 ppm) and deoxyribose (6.7 mM). An equivalent volume of TCA (3% w/v) aqueous solution was added after 20 min incubation at room temperature to stop the Fenton reaction. The solution was then filtered through 0.2 μm membrane to remove the FLG or GO. Part of the filtrate (2 mL) was moved into a test tube and TBA (1 mL, 1%) aqueous dispersion was added. The test tube was kept in a boiling water bath for 15 min to develop the pink chromogen. The tube was then cooled down in another water bath at room temperature for 10 min. The solution was then moved into cuvette and absorbance by the pink chromogen at 532 nm was then recorded with a JASCO V-630 UV/Vis Spectrometer. To rule out adsorptive artifacts, the ability of FLG to physically adsorb deoxyribose was characterized by pre-incubating deoxyribose solutions with FLG for 10 min and then removing the solids with filtration. The filtrate was then incubated with the Fenton reagents and the pink chromogen was developed and measured in the same way. To further test for artifacts involving MDA adsorption, the same Fenton reaction was carried out without FLG for 20 min after which TCA and FLG were added. After a further 10 min incubation, the FLG was filtered after and the filtrates treated as above to develop and measure the pink chromogen.

4.2.3.9 Lipid peroxidation

Linoleic acid oxidation is used as a model in this study. The oxidation is initiated by an azo initiator AAPH.⁴³ Fresh linoleic acid (150 μL) was dispersed in PBS (150 μL , 50 mM, pH 7.4) with SDS (10 mM) as surfactant. The solution was then diluted 20 times and added tested

materials (200 ppm final concentration). Oxidation is initiated by the addition of AAPH (2.4 mM final concentration) into the above solution preheated in 40 °C water bath. The progress of oxidation was monitored by taking a small sample of the solution and measuring the amount of lipid peroxides, which is determined by boiling the sample with TBA (3% w/v) for 15 min and comparing the absorbance at 531 nm.⁴⁴ Higher absorbance indicates lipid oxidized to higher extent.

4.2.3.10 TiO₂ encapsulation by graphene for ROS suppression

Here 0.01 mM Rhodamine B (RhB) was used to monitor the production of $\cdot\text{OH}$ -induced by UV irradiation (365 nm at $\sim 10 \text{ mW cm}^{-2}$) and the relative quenching ability of GO added. Water is transparent to the wavelength used here.⁴⁵ The suspension containing GO (20 ppm) and TiO₂ (80 ppm) looks uniform without agglomeration in PBS (pH 7.4).⁴⁶ An intrinsic contact between graphene based materials and TiO₂ is achieved. We used a method similar to a previous study by our group to produce TiO₂ graphene nanosacks.⁴⁷ Nanosacks (100 ppm, $\sim 80\%$ TiO₂ and $\sim 20\%$ graphene-based materials) were used for comparison. The mixture of dye, TiO₂ (and GO) in buffer was left for 30 min in darkness under continuous stirring with rotator for physisorption before exposure. After exposed to UV light with a distance of $\sim 5 \text{ cm}$ between irradiation source and the solution under continuous stirring for various length of time the solid materials were removed through filtration and the absorbance of RhB was recorded.

4.3 Results and discussion

4.3.1 Characterization

Figure 4.1 summarizes the properties of the graphene-based materials synthesized and used in this study. The GO has typical lateral dimensions from 1–5 μm (Figure 4.1B) and is primarily in monolayer form (Figure 4.1C), which exhibits $\sim 2600 \text{ m}^2 \text{ g}^{-1}$ surface area when fully dispersed in aqueous media. In contrast, the FLG and rGO materials are multilayer structures that were obtained/fabricated as dry powders that must be dispersed in the aqueous test media. Vapor adsorption methods provide information on surface area and pore structure of these graphene-based powders (Figure 4.1D), which are lower ($663\text{--}713 \text{ m}^2 \text{ g}^{-1}$) than the monolayer surface area due to face-to-face association of individual layers (or incomplete exfoliation in some cases). The surface area of rGO powders measured by N_2 probe is only $10 \text{ m}^2 \text{ g}^{-1}$ (data not shown) but $680 \text{ m}^2 \text{ g}^{-1}$ when measured by CO_2 probe. This is a well-known effect in carbon materials ($\text{Area}_{\text{nitrogen}} \ll \text{Area}_{\text{carbon-dioxide}}$) that indicates the presence of super-micropores near the molecular diameter of N_2 (0.3–0.84 nm) that are not accessible to the N_2 probe at the low temperature of the experiment (77 K) due to diffusional limitations but are accessible to CO_2 (a similarly sized molecule) at the higher temperatures used to obtain those isotherms (273 K).⁴⁸ The presence of these ultrafine micropores capable of molecular sieving has been observed previously in rGO films.⁴⁹ Raman characterization (Figure 4.1A) shows lower defect densities on FLG, as expected for this material that has not been subjected to permanganate-mediated basal surface oxidation.

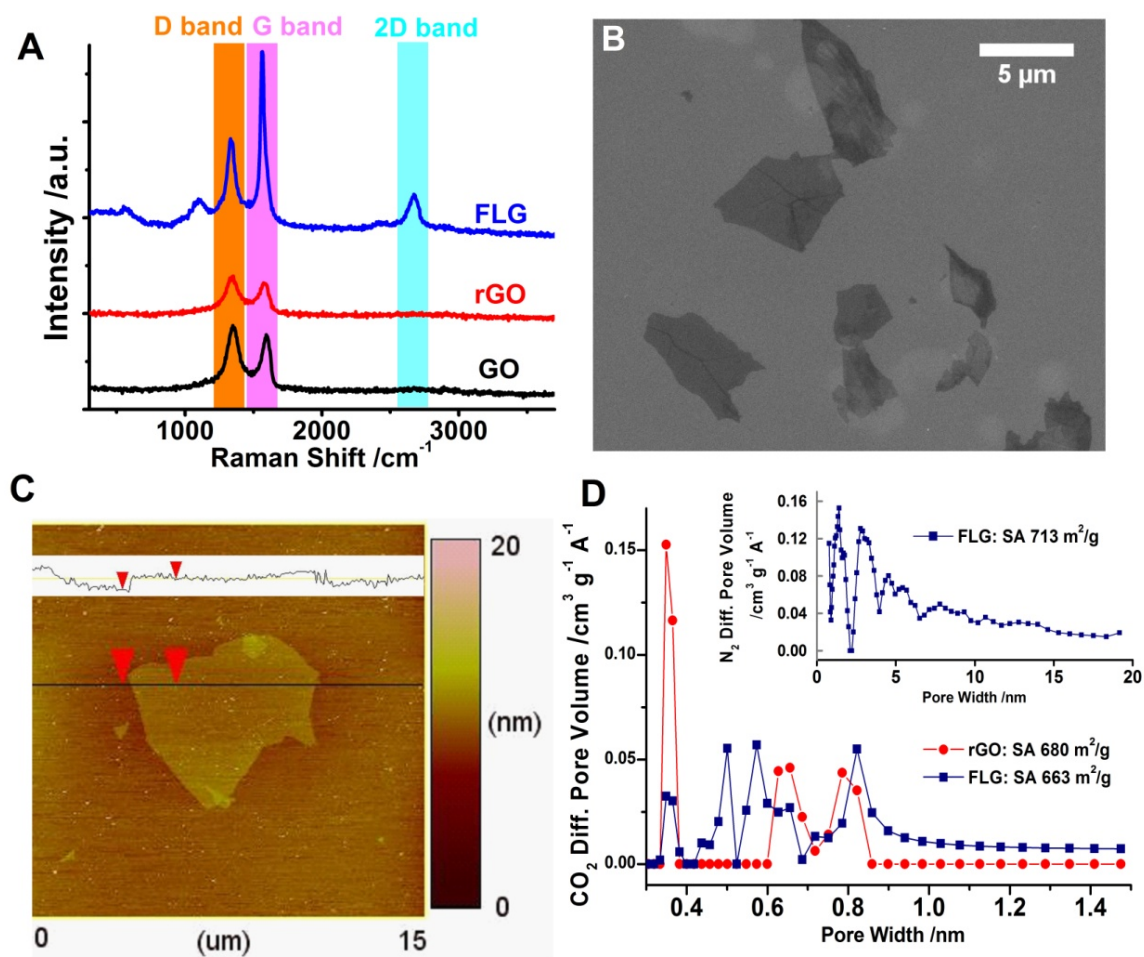


Figure 4.1 GO based materials characterization.

(A) Raman spectra; (B) Typical SEM image showing 1-5 μm nominal lateral dimension for GO sheets on silicon substrate; (C) AFM image of typical GO sheet. Line scan shows 1.04 nm step height indicating monolayer GO; D: Differential pore size distributions (Diff. PSD) for rGO and FLG as bulk dry powders as probed by CO₂ at 273 K (main plot) and N₂ at 77 K (inset). The total surface areas near 700 m²g⁻¹ indicate that the materials are composed of multilayer stacks of approximately 3-4 layers (2600/700). The N₂ Diff. PSD show characteristic modes with a periodic spacing of 1-2 nm, which has been interpreted as the pores formed by irregular stacking of 1-2 nm plate-like structures

4.3.2 Hydroxyl radical scavenging

Multiple assays were employed to characterize the ability of graphene-based materials to scavenge ·OH, which is a highly reactive radical implicated in a wide range of undesired low-temperature oxidation processes and biochemical reactions. Figure 4.2 shows the

ability of (monolayer) GO in suspension to protect organic molecules in solution from $\cdot\text{OH}$ oxidation. Here the $\cdot\text{OH}$ is generated by Fenton chemistry ($\text{Fe}^{2+} + \text{H}_2\text{O}_2 \rightarrow \cdot\text{OH} + \text{OH}^- + \text{Fe}^{3+}$), and the oxidizable target molecules are tracked by UV-vis. spectroscopy (for the case of phenol red, Figure 4.2A) or by EPR (for DMPO, which forms the DMPO-OH radical adduct with its characteristic EPR spectrum shown in Figure 4.2B). The protective effect of GO in both cases is dose-dependent and quite pronounced above $100 \mu\text{g mL}^{-1}$ (100 ppm).

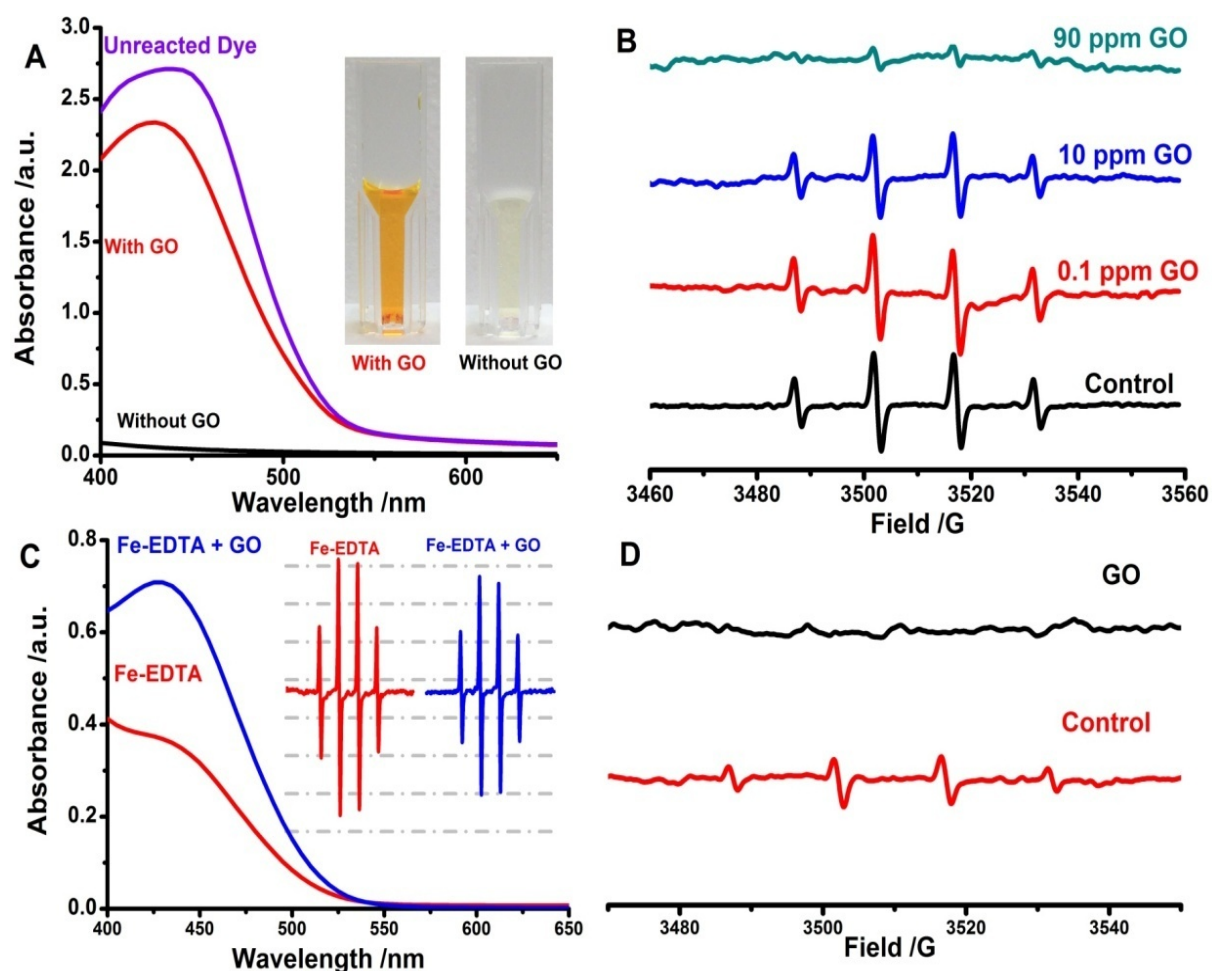


Figure 4.2 GO as a hydroxyl-radical scavenger.

(A) Addition of 100 ppm GO protects phenol red from oxidation by Fenton-generated $\cdot\text{OH}$ over 8 hrs. (B) Addition of GO protects the spin-trap DMPO from oxidation by Fenton-generated $\cdot\text{OH}$ detected by EPR spectroscopy. The protective effect is concentration dependent over the range 0.1-90 ppm GO. (C) Both phenol red monitoring and EPR were used to test if the GO-mediated protective effect is due to radical scavenging or Fenton suppression by Fe-binding. The protective

effect of GO is still present when GO-Fe binding is suppressed by EDTA, confirming the importance of radical scavenging. (D) GO also protects DMPO when $\cdot\text{OH}$ is generated sonochemically, in a non-Fenton assay, providing additional evidence for true radical scavenging. All experiments were done in PBS (pH 3.5).

We considered the possibility that the observed effect was due to inhibition of the Fenton reaction by iron complexation at oxygen-containing functional groups on GO rather than radical scavenging, and carried out two control experiments to evaluate this possibility. Figure 4.2C shows that the GO protective effect is still observed when an excess of ethylenediamine tetra acetic acid (EDTA) is added, whose strong chelation ability prevents iron complexation with GO. (Separate experiments involving iron analysis by inductively coupled plasma-atomic emission spectroscopy (ICP-ES) show that GO does bind iron in these solutions, but not in the presence of the much stronger complexing agent EDTA – see ESI). The EDTA-Fe complex retains some redox activity, which leads to phenol red oxidation and DMPO adduct formation, and these oxidation processes are still inhibited by GO, confirming the presence of the radical scavenging mechanism. Further, Figure 4.2D shows that GO is still protective when $\cdot\text{OH}$ is generated by sonochemistry, which involves homolytic cleavage of H_2O_2 ⁵¹⁻⁵² by a mechanism that does not involve iron. Another possible artifact here would be the direct reaction between GO and H_2O_2 used in the Fenton-generation of $\cdot\text{OH}$, but this reaction was found to be slow by direct measurement of H_2O_2 (see ESI) and through a later assay (Figure 4.6A). Overall, it is clear that dispersed monolayer GO at 100 ppm concentration is quite active as an $\cdot\text{OH}$ scavenger.

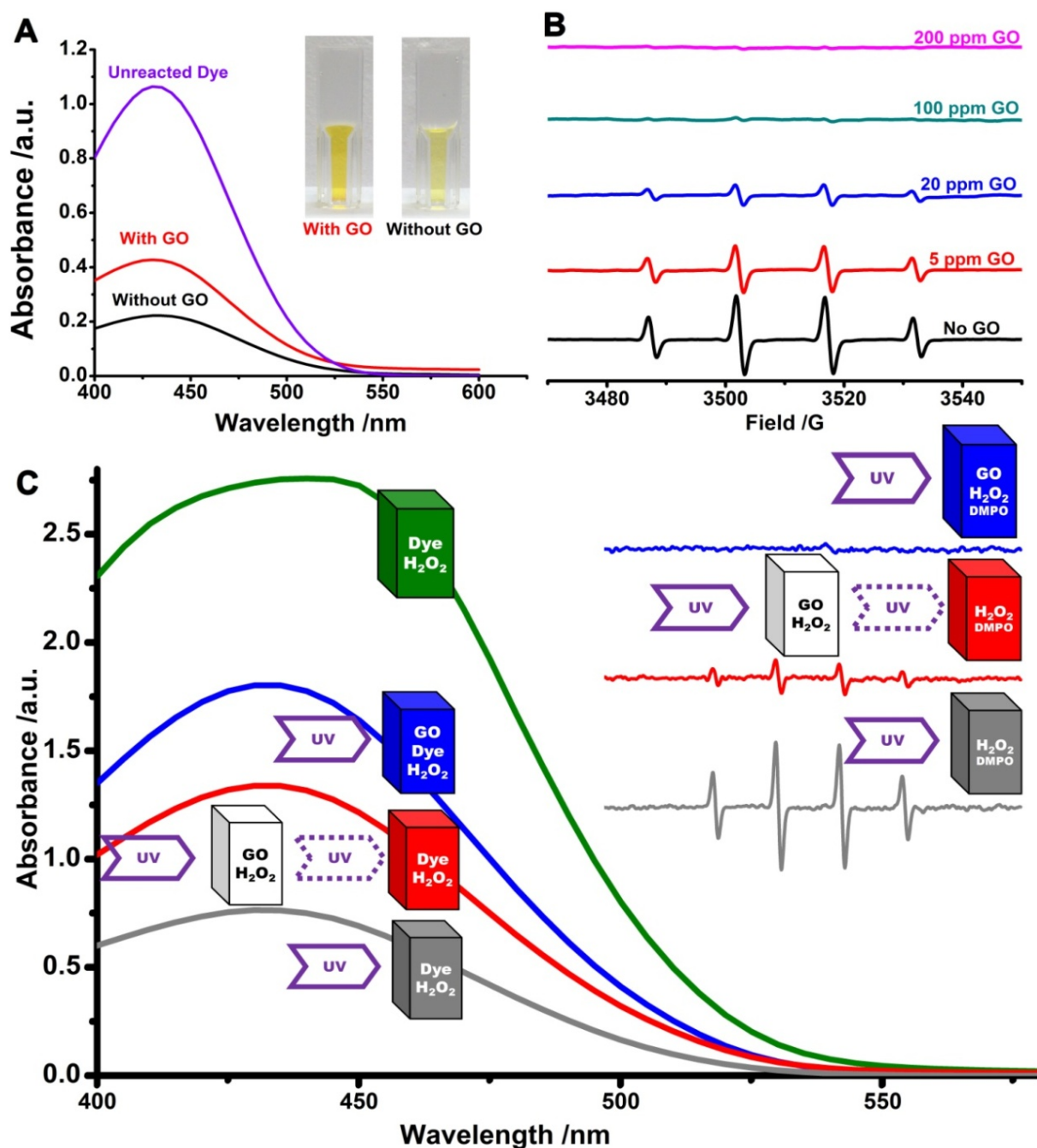


Figure 4.3 GO antioxidant function in systems with $\cdot\text{OH}$ generation by UV-irradiation of hydrogen peroxide.

(A) GO (200 ppm) retards phenol red oxidation, (B) GO (200 ppm) suppresses DMPO- OH radical adduct formation; (C) Mechanism - additional experiments to separate the effects of radical scavenging and UV adsorption by GO. Placing GO in the UV beam path but physically separated from the dye reveals the contribution of UV absorption to the total protective effect of GO. Results from both dye monitoring (main plot, 200 ppm GO) and EPR (inset, 100 ppm GO) show that the protective effect of GO in these systems is a combination of UV absorption and radical scavenging.

Figure 4.3 characterizes the behavior of GO in systems where $\cdot\text{OH}$ is generated by water photolysis rather than Fenton chemistry. Here again, suspended monolayer GO shows some protective effect for phenol red (Figure 4.3A) and for DMPO (Figure 4.3B). These effects cannot be related to iron binding, but may involve $\cdot\text{OH}$ scavenging or UV absorption that inhibits the original $\cdot\text{OH}$ generation. Figure 4.3C shows the results of special control experiments to determine the separate contributions of UV absorption and $\cdot\text{OH}$ scavenging. The red curves (second from bottom) were generated by placing the GO in a separate cuvette - isolated from the target molecule, but in the beam path where UV absorption can occur but not scavenging in a manner that could protect the phenol red (main plot) or the DMPO (inset). These results show that the antioxidant effect of GO here is a combination of UV absorption and $\cdot\text{OH}$ scavenging. The UV absorption is a type of preventative antioxidation mechanism, and because this absorption is the result of integration along the total beam path, it should be more important in thicker systems (tissues, bulk materials or thick films) and less important in thin film systems according to Beer-Lambert law.

An important goal of this work is to explore the relative antioxidant activity of different members of the graphene material family as a guide to material selection and design.

We attempted to use all three materials (GO, rGO, FLG) in all assays, but found that the hydrophobic varieties (rGO, FLG) produced adsorptive artifacts, especially in those assays involving dye destruction, due to the well-known affinity of pristine carbon surfaces for conjugated dyes. We therefore focused on GO in most assays, but devised special experiments to compare the three materials in a reliable manner free of artifacts. Figure 4.4A compares the $\cdot\text{OH}$ scavenging activities of GO, rGO, FLG, and reference antioxidants using the DMPO spin trap protection assay with Fenton-generated $\cdot\text{OH}$ and EPR detection of DMPO-OH as above. At 200 ppm, all the graphene-based materials are active scavengers and

are more active than the reference antioxidants mannitol and the fullerene derivative pyrrolidine tris-acid C₆₀ (pC60), which is a commercially available water-soluble fullerene derivative reported to show antioxidant activity.²⁴ Ascorbic acid is seen to be a pro-oxidant under these conditions, as has been observed previously,⁵³ and attributed to its activity in reducing Fe³⁺ to Fe²⁺ for further Fenton reaction with peroxide^{11, 54}, which accelerates ·OH generation in this assay.

The antioxidant activity of the graphene-based materials toward ·OH falls in the order FLG > rGO > GO. Because monolayer GO has by far the highest surface area, this rank order indicates a much higher area-specific (intrinsic) activity for the pristine or reduced graphene forms (FLG, rGO), an observation that provides insight into the mechanism of antioxidation (see Discussion). The left two bars in Figure 4.4B shows that FLG is more active than GO in protecting deoxyribose from ·OH attack. We were concerned about artifacts, since the pristine hydrophobic surfaces of the FLG samples can cause significant physical adsorption of dyes and reagents in biological fluids and assays.⁵⁵ We therefore carried out an additional assay with deoxyribose as a very soluble hydrophilic target molecule that is not depleted by physical adsorption under these conditions (the right two bars in Figure 4.4B). Here too FLG is more active indicating that it is an intrinsically better ·OH scavenger than GO. Additional controls indicate that the EPR effects in Figure 4.4A are not due to FLG interacting with DMPO or its OH adduct (see ESI).

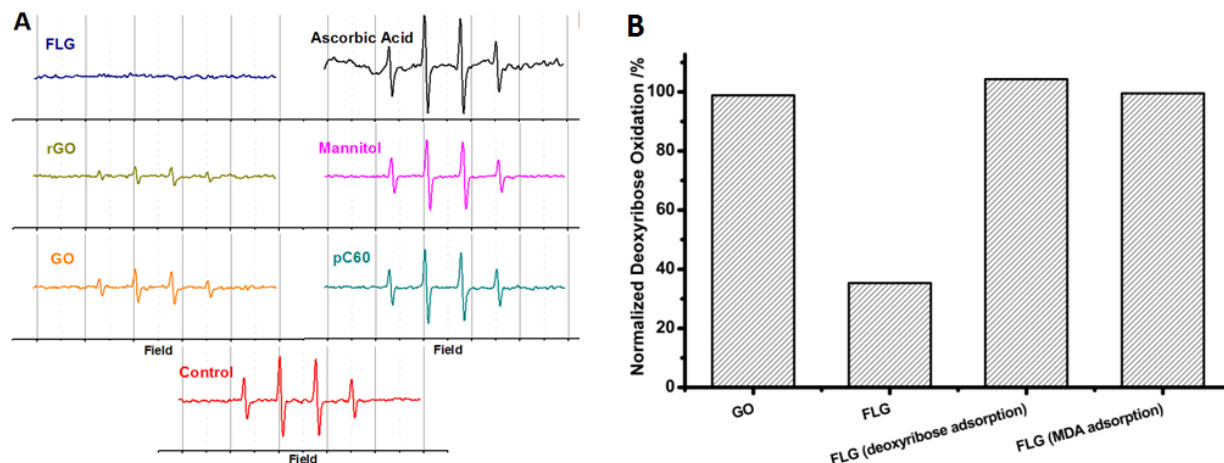


Figure 4.4 Comparison of different graphene-based materials as ·OH scavengers.

(A) EPR spectra of DMPO-·OH adduct formed through 20 min Fenton reaction- in the presence of 200 ppm graphene-based and reference materials. Radical scavenging potency follows rank order: FLG > rGO > GO > mannitol > pC60. Ascorbic acid is a pro-oxidant under these conditions. (B) MDA assay showing 100 ppm FLG is more active than 100 ppm GO in protection of 100 mM deoxyribose from Fenton-generated ·OH oxidation. The right-hand two bars show control experiments that rule out adsorptive artifacts in the FLG experiment. When FLG is added and removed before the oxidation step (third bar), there is no effect (which rules out physical adsorption of deoxyribose). When FLG is added and removed after the oxidation (fourth bar), there is also no effect (which rules out physical adsorption of the deoxyribose oxidation products)

4.3.3 Other radical and oxidant interactions

Superoxide anion, $O_2^{\cdot-}$, is another important oxygen-centered radical species that forms through the one-electron reduction of molecular oxygen, and is scavenged in biological system by the superoxide dismutase family of antioxidant enzymes that catalyze its destruction through disproportionation to H_2O_2 and O_2 .⁵⁶ Figure 4.5 shows the effects of GO on superoxide concentrations following $O_2^{\cdot-}$ generation by non-enzymatic (A) and enzymatic (B) routes. The assay monitors the superoxide-mediated conversion of nitroblue tetrazolium chloride (NBT) to the blue chromagen formazan, which has a characteristic absorption peak at 560 nm.³⁸ Figure 4.5A shows that GO shows a modest scavenging effect against superoxide compared to ascorbic acid and pC60, which both have been reported as

strong $O_2^{\cdot-}$ scavengers.^{24, 57-59} Because the activity is seen in both the enzymatic and non-enzymatic systems, the results cannot be attributed to GO-mediated enzyme deactivation.

In addition to the protection-based, three-molecule (oxidant, target, antioxidant) assays described above, antioxidant activity can also be assessed by measuring the direct reaction rates of candidate antioxidant molecules (here graphene-based materials) with model free radicals of sufficient stability for use in laboratory assays. Figure 4.5C shows results for the standard radical DPPH.^{34, 39, 60} and Figure 4.5D for ABTS.^{+,41} GO shows weak activity toward DPPH $\cdot$ in comparison to the known reference antioxidants pC60 and ascorbate.⁷ Similarly, GO shows weak but measureable activity toward ABTS $^{+}$ relative to known antioxidants ascorbate, pC60, and Trolox.^{41, 61} Because the quenching of DPPH $\cdot$ and ABTS $^{+}$ typically occur through hydrogen donation⁴⁰⁻⁴¹ the weak activity of GO here suggests it is a poor H-donor.

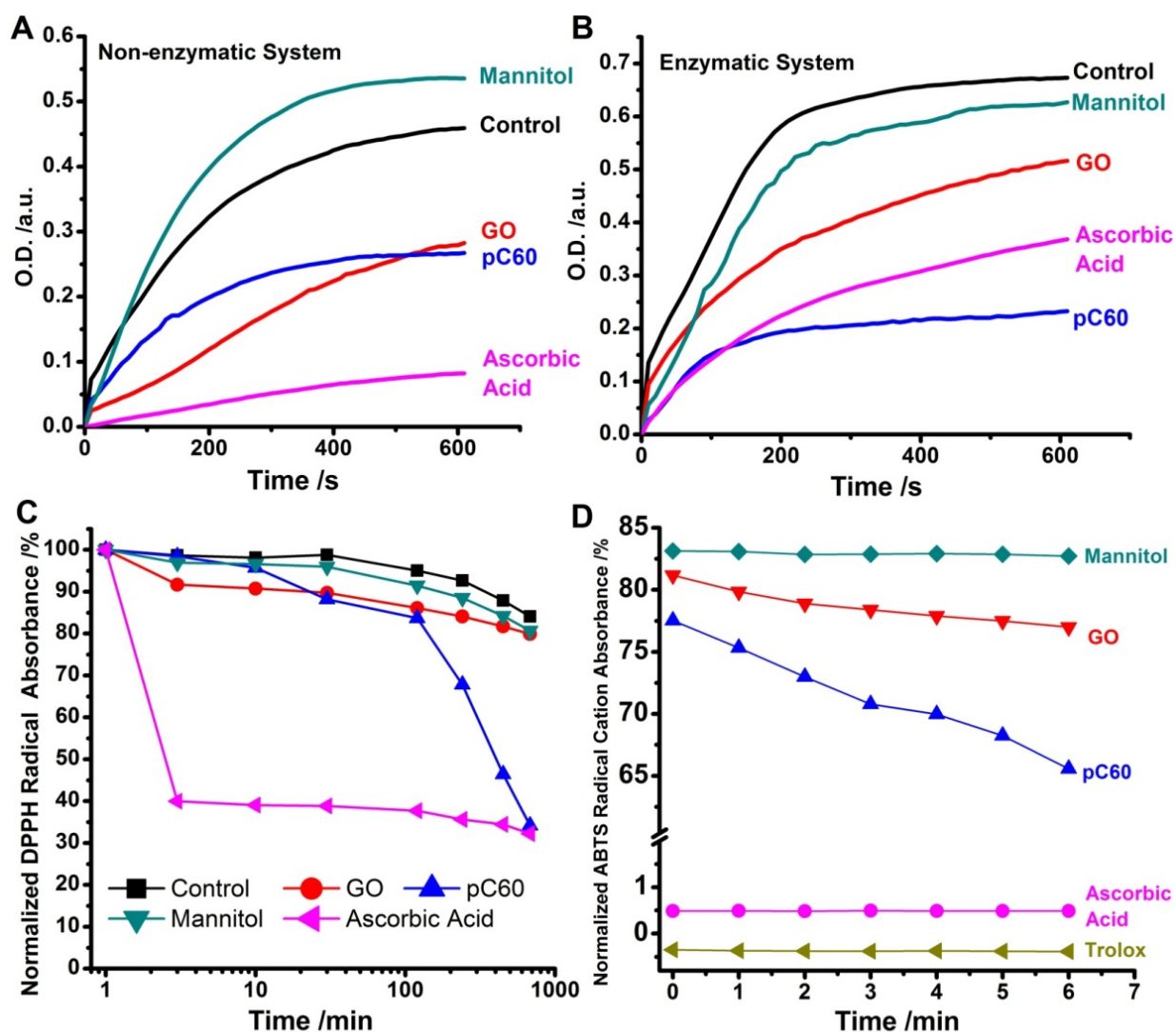


Figure 4.5 GO interactions with other radical species.

(A,B) superoxide radical scavenging by GO and reference antioxidants (83 ppm) using nonenzymatic (A) and enzymatic (B) methods of superoxide generation. Superoxide was monitored for 600 s by measuring the absorbance of superoxide-reduced NBT at 560 nm. GO shows modest scavenging activity in this assay. C, D: Interaction of GO and reference antioxidants (100 ppm) with the model stable free radicals DPPH $\cdot$ (C) and ABTS $^{+}$ radical cation (D). GO shows weak activity relative to the known H-donor antioxidants ascorbic acid and Trolox.

Glutathione (GSH) is a key endogenous antioxidant, whose depletion is widely used as a measure of oxidative stress in biological systems.^{56, 62-63} GSH oxidation can be catalyzed by carbon nanotubes¹⁴ and an acellular GSH oxidation assay has been proposed as a measure of pro-oxidant potential for carbon nanomaterials.¹⁴⁻¹⁵ GSH reacts quickly with hydrogen

peroxide ¹⁴ and here we carried out experiments to see if GO could inhibit that oxidation and protect GSH. Figure 4.6A shows that GO has no measurable ability to protect GSH from H₂O₂. Figure 4.6B shows that the presence of GO caused a slight delay in lipid peroxidation initiated by the azo free-radical initiator 2,2'-azobis(2-amidinopropane) hydrochloride (AAPH) which decomposes by a first order kinetics. Lipid peroxidation is a major cause of food and drug product spoilage and has been implicated in human disease.⁴ Here GO shows a weaker antioxidant activity than Trolox, an analog of vitamin E used as a standard reference antioxidant for lipid peroxidation.

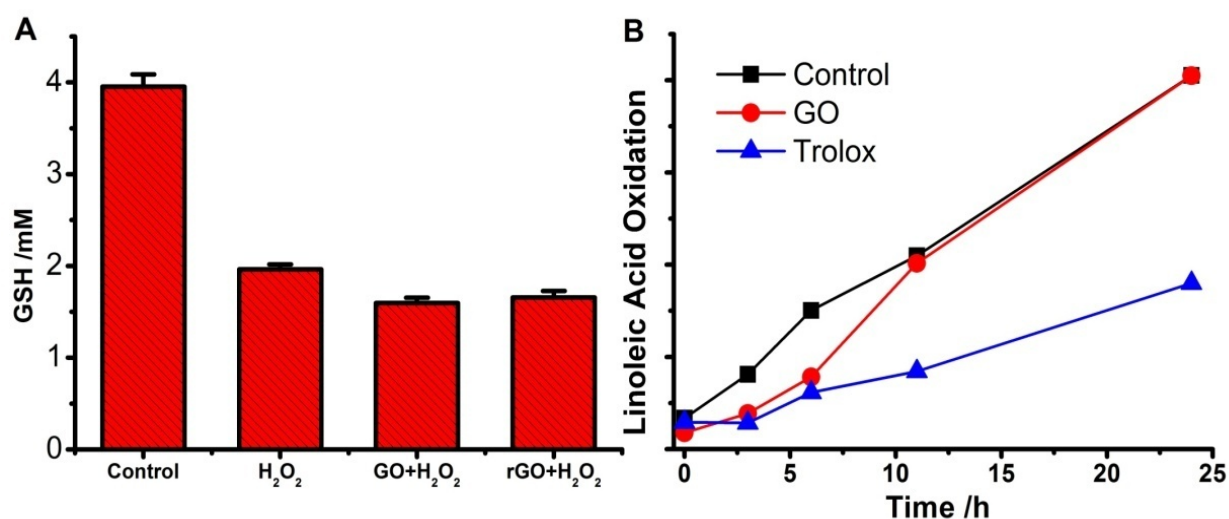


Figure 4.6 Graphene material activity against hydrogen peroxide and lipid peroxyl radicals

(A) GO and rGO (100 ppm) shows no protection on 4 mM GSH against oxidation by 1 mM H₂O₂. (B) GO (200 ppm) shows weak protection on linoleic acid against oxidation.

4.3.4 Discussion

Figures 4.2-6 provide the first systematic data on the antioxidant behaviors of graphene-based materials. A number of clear patterns are seen: first they are active

against $\cdot\text{OH}$ in multiple independent assays, and when present in a well-dispersed state at ~ 100 ppm can protect dissolved organic dyes and spin traps from $\cdot\text{OH}$ oxidation. They show modest activity against superoxide, but are relatively inactive toward hydrogen peroxide, lipid peroxy radicals, and two stable free radicals (DPPH $\cdot$, ABTS $^{\cdot+}$) that are often used as standard probes to assess the activity of molecular antioxidants. The graphene-based materials show OH radical scavenging activities in the order $\text{FLG} > \text{rGO} > \text{GO}$, which is in inverse order to their total surface area.

To what extent can this pattern of antioxidant activity be related to the structures of GO or pristine FLG? First we note that graphene materials may also have pro-oxidant activities, and the observed ROS inhibition here is, in principle, the net effect of ROS scavenging and generation, though the high concentrations of externally generated ROS may be expected to overwhelm the material-dependent ROS. We have carried out studies of oxidant production from this set of materials (see ESI), and the order of activity is also $\text{FLG} > \text{rGO} > \text{GO}$, so pro-oxidant contributions cannot explain the trends seen here for anti-oxidant activity.

The ranking $\text{FLG} > \text{rGO} > \text{GO}$ suggests that the primary active sites are associated with the pristine graphenic network, rather than oxygen-containing functional groups. We note that the activity of oxygen-containing groups may be pH-dependent, but the high activity of FLG relative to GO is seen at both pH3 and pH7 in separate assays. Most molecular antioxidants that show activity toward DPPH $\cdot$ and ABTS $^{\cdot+}$ are hydrogen donors of the form AH that act through: $\text{AH} + \text{R}\cdot \rightarrow \text{RH} + \text{A}\cdot$, where the new radical $\text{A}\cdot$ is sufficiently stable not to propagate the free-radical chain reaction. Many H-donor antioxidants for $\cdot\text{OH}$ scavenging act through R-O-H groups where the O-H bond dissociation energy (BDE) is low enough for sufficient reactivity.^{32, 64} It is interesting that GO is seen here to be a weak antioxidant in these standard assays, and is thus a poor H-donor antioxidant despite its high hydroxyl

content compared with rGO and FLG as shown by the FTIR spectra in ESI (OH/C ratio ~ 0.25 as reported in literature).⁶⁵

We believe the weak activity of GO as a donor antioxidant is consistent with its chemical structure. Many antioxidants are phenolic compounds,¹ whose radical forms following H-donation are stabilized by resonance structures,⁶⁶ in which the unpaired electron can reside on the oxygen atom, or on ortho or para carbons on the adjacent aromatic ring.⁶⁷ GO has many hydroxyl groups and also extensive aromaticity in the form of pristine sp^2 -carbon domains between oxidized regions, but is not highly active as an H-donor. Most accepted models for GO structure place the OH groups out-of-plane at basal sites, where oxidation of C=C double bonds has created local sp^3 sites that do not offer the adjacent conjugated structure necessary for radical resonant stabilization. Such basal OH groups on GO are not expected to show strong antioxidant activity.

In the absence of basal defects in GO, the only source of phenolic groups (aromatic hydroxyl sites) would be at the sheet periphery. Such sites are very few, however, for the GO sheets of microscale lateral dimension ($L \sim 1 \mu\text{m}$) used here and in most other studies. The ratio of basal-to-edge sites scales as $L_{\text{sheet}}/L_{\text{C-C-bond}}$, which is of order 10^3 for microscale sheets. Even if phenolic OH groups were to cover the entire GO edge, at 1:1000 edge-to-basal-site ratio, phenols cannot realistically contribute a significant fraction of the total hydroxyl sites in GO. Most OH in GO must be non-phenolic, and this is consistent with the low H-donor antioxidant activity observed here.

Figure 4.7A summarizes the behaviors observed in this study. Graphene-based materials are effective scavengers hydroxyl radical, and show some activity toward superoxide. The primary radical scavenging sites are believed to be associated with the pristine sp^2 -carbon domains, which act through adduct formation²⁸ or electron transfer.

Graphene-based materials may also act as preventative antioxidants through UV absorption during photochemical ROS formation. Graphene-based materials including GO are weak H-donor antioxidants, which together with the fact that they are large non-diffusible species limits their use to certain applications in oxidation protection. Graphene-based sheet-like materials of microscale lateral dimension can be thought of as stationary, passive scavengers of the more reactive species, such as $\cdot\text{OH}$, and to be effective in this data, must be present at concentrations on the same order as the target molecules they protect. In some terminologies, materials in this class are referred to oxidation retarders rather than true antioxidants,⁶⁸ the latter term being reserved for materials that are effective at much lower concentrations. This pattern of activity suggests two possible antioxidant applications for graphene-based materials:

(1) as fillers or additives in a continuous organic matrix phase, where their ultrahigh surface area provides good site availability and large capacity for $\cdot\text{OH}$ or $\cdot\text{O}_2^-$ scavenging. These applications will require high concentrations of the carbon material,³ in a well-dispersed state, and are thus likely of most interest when the graphene material also provides a second function as a conductive filler or mechanical reinforcement.⁶⁹

(2) as planar films or encapsulation shells where the graphene material serves both as a chemical antioxidant and a physical barrier to oxidant transport to achieve a high degree of oxidation protection. This class of application makes use of the unique 2D geometry of graphene.

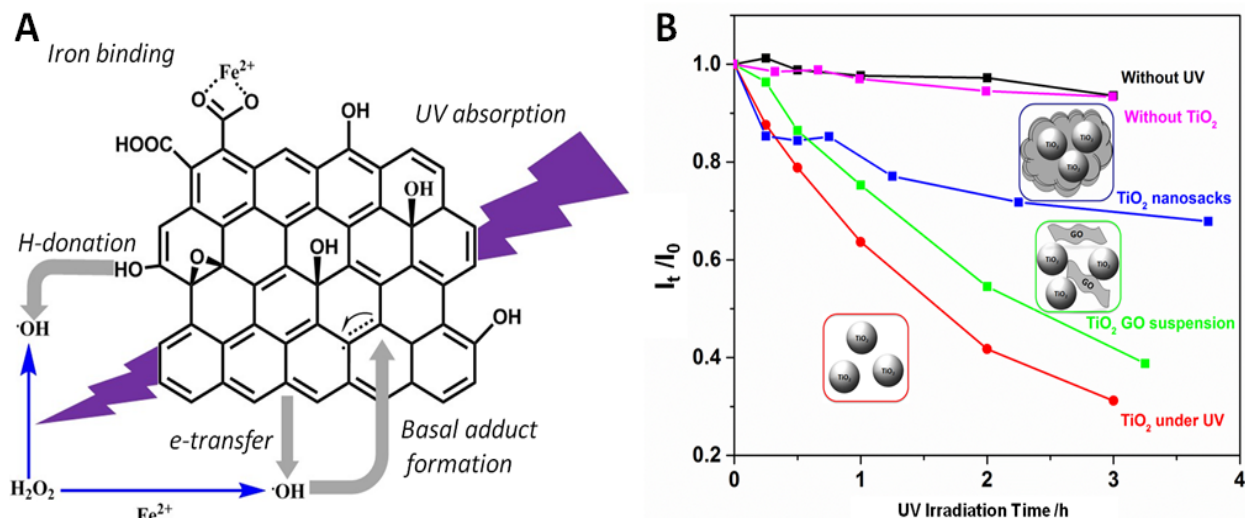


Figure 4.7 Antioxidant mechanisms and example application.

(A) Overview of relevant antioxidant mechanisms (UV absorption, iron binding, $\cdot\text{OH}$ adduct formation on sp^2 -carbon sites; electron transfer, and hydrogen donation). This study shows that the primary antioxidant activity for graphene-based materials is against $\cdot\text{OH}$ and is associated with pristine sp^2 -carbon domains. Hydrogen donation activity is limited by the low population of phenolic OH, which are found on edge but not basal sites. (B) Example application of graphene-based oxidation protection. TiO_2 -mediated photocatalytic destruction of Rhodamine B dye in the presence and absence of graphene-based materials when exposed to 365 nm long-wavelength UV. Addition of GO (final concentration 20 ppm) in suspension slightly inhibits dye destruction. Encapsulation of TiO_2 in graphene nanosacks (0.1 mg mL^{-1} nanosacks with $\sim 80 \text{ wt\%}$ TiO_2 and $\sim 20 \text{ wt\%}$ rGO) significantly inhibits dye oxidation. ROS produced at TiO_2 surfaces are scavenged by internal rGO surfaces prior to release and dye oxidation.

As a first demonstration of application type (2) we explored the potential for graphene encapsulation to suppress the release of ROS from TiO_2 nanoparticle surfaces. TiO_2 nanoparticles are widely used in exterior coatings, paints and sunscreens to scatter and absorb potentially damaging ultraviolet radiation (290-400 nm) in sunlight.⁷⁰ Ultraviolet photon absorption produces electron-hole pairs in TiO_2 that lead to ROS formation through surface reactions,⁷¹ which in turn causes oxidative damage in the paint, coating, or adjacent skin tissue. Organic and inorganic coatings have been used to suppress ROS formation and protect the product or tissue.⁷²⁻⁷³ The present results suggest that graphene-based materials may be effective in this role both through UV adsorption and radical quenching if a suitable TiO_2 -graphene hybrid architecture can be designed.

One possible architecture involves graphene encapsulation, which has been demonstrated through electrostatic wrapping⁷⁴⁻⁷⁵ or through continuous aerosol microdroplet drying to form filled graphene nanosacks.^{47, 76-77} An important pathway for TiO₂-mediated oxidative damage involves ·OH production according to: OH⁺ + hole⁺ → ·OH⁷⁸ followed by ·OH diffusion from the surface to attack adjacent molecular targets. In a TiO₂-filled graphene nanosack, these surface hole sites would exist only inside the folded sack, and the ·OH produced would have to diffuse through the porous interior and around internal graphene structures to reach surrounding tissue. We hypothesized that the high reactivity of ·OH toward graphenic surfaces would lead to quenching before radical release, and that graphene sack encapsulation would be effective at mitigating TiO₂ nanoparticle-induced oxidative damage.

Figure 4.7B shows the effect of graphene-based materials on TiO₂-induced photocatalytic dye destruction. TiO₂ nanoparticles alone catalyze the destruction of most of the dye (Rhodamine B) within 3 hrs, and addition of GO in co-suspension has a slight inhibitory effect under these conditions. Much more significant inhibition is seen when all of the TiO₂ particles are encapsulated in folded graphene nanosacks, which are known to be multilayer (3-10) rGO structures whose irregular folding allows rapid diffusional exchange of solutes in aqueous phases,⁴⁷ here oxygen and ROS. The overall antioxidant function seen in Figure 4.7B is believed to be a combination of UV absorption, ROS scavenging in the graphene sack interior, and possible passivation of a portion of the TiO₂ surfaces by close contact with internal conforming graphene structures (see SI) in a manner analogous to the passivation of silica by carbon black following co-grinding to produce intimate contact.⁷⁹

It is interesting that some other studies report the opposite trend - *increases* in TiO₂ photochemical activity following hybridization with graphene.⁸⁰⁻⁸¹ These prior studies

fabricate TiO_2 nanostructures bound to planar graphene sheets and attribute the enhancement to electron transfer from TiO_2 to graphene, which reduces electron-hole recombination and improves the photochemical efficiency. This same effect may be present in the data in Figure 4.7B, but is overwhelmed by the antioxidant function in the nanosack configuration, and the net result is significant inhibition of ROS and oxidative damage. We believe this is a unique behavior of the sack-cargo architecture, which places the oxidizing sites (holes) entirely inside a porous graphenic structure that provides numerous sites for radical scavenging before ROS species can be released to the surrounding medium. This simple demonstration suggests that graphene sack encapsulation may be useful for managing the environmental and health risks associated with some nanoparticle-based technologies.

4.4 Conclusions

Here we show that graphene oxide exhibits significant antioxidant activity in the form of hydroxyl and superoxide radical scavenging, and can protect a variety of biomolecular target molecules from oxidation. Thermally reduced rGO and few-layer graphene are more active than GO despite its lower surface area, indicating that the main scavenging activity is associated with pristine sp^2 carbon domains on basal surfaces rather than H-donation from hydroxyl or hydroquinone groups. In experiments involving UV-induced $\cdot\text{OH}$ generation, the protective effect is a combination of UV absorption by graphene and radical scavenging. The combination of radical scavenging, UV absorption, ultrahigh surface area, and elasticity make GO based materials promising for selected antioxidant applications as dispersed phase fillers, or as conformal encapsulating agents that can mitigate ROS production and toxicity from redox-active nanoparticle cargos.

4.5 References

1. Ho, C.-T., Phenolic Compounds in Food. In *Phenolic Compounds in Food and Their Effects on Health II*, American Chemical Society: 1992; Vol. 507, pp 2-7.
2. Antolovich, M.; Prenzler, P. D.; Patsalides, E.; McDonald, S.; Robards, K., Methods for testing antioxidant activity. *Analyst* 2002, 127 (1), 183-198.
3. Watts, P. C. P.; Fearon, P. K.; Hsu, W. K.; Billingham, N. C.; Kroto, H. W.; Walton, D. R. M., Carbon nanotubes as polymer antioxidants. *J. Mater. Chem.* 2003, 13 (3), 491-495.
4. Yin, H. Y.; Xu, L. B.; Porter, N. A., Free Radical Lipid Peroxidation: Mechanisms and Analysis. *Chem. Rev.* 2011, 111 (10), 5944-5972.
5. Lipinski, B., Hydroxyl Radical and Its Scavengers in Health and Disease. *Oxid. Med. Cell. Longev.* 2011, 2011, 809696.
6. Ohshima, H.; Yoshie, Y.; Auriol, S.; Gilibert, I., Antioxidant and pro-oxidant actions of flavonoids: Effects on DNA damage induced by nitric oxide, peroxynitrite and nitroxyl anion. *Free Radical Biol. Med.* 1998, 25 (9), 1057-1065.
7. Silva, B. M.; Andrade, P. B.; Valentao, P.; Ferreres, F.; Seabra, R. M.; Ferreira, M. A., Quince (*Cydonia oblonga* Miller) fruit (pulp, peel, and seed) and Jam: antioxidant activity. *J. Agric. Food. Chem.* 2004, 52 (15), 4705-12.
8. Pyrzynska, K.; Pekal, A., Application of free radical diphenylpicrylhydrazyl (DPPH) to estimate the antioxidant capacity of food samples. *Analytical Methods* 2013, 5 (17), 4288-4295.
9. Pietta, P. G., Flavonoids as antioxidants. *J. Nat. Prod.* 2000, 63 (7), 1035-1042.
10. Christenson, E. M.; Anderson, J. M.; Hiltner, A., Antioxidant inhibition of poly(carbonate urethane) in vivo biodegradation. *Journal of Biomedical Materials Research Part A* 2006, 76A (3), 480-490.

11. Buettner, G. R.; Jurkiewicz, B. A., Catalytic metals, ascorbate and free radicals: Combinations to avoid. *Radiat. Res.* 1996, *145* (5), 532-541.
12. Almeida, E. S.; Portela, F. M.; Sousa, R. M. F.; Daniel, D.; Terrones, M. G. H.; Richter, E. M.; Munoz, R. A. A., Behaviour of the antioxidant tert-butylhydroquinone on the storage stability and corrosive character of biodiesel. *Fuel* 2011, *90* (11), 3480-3484.
13. Sen, C. K.; Khanna, S.; Gordillo, G.; Bagchi, D.; Bagchi, M.; Roy, S., Oxygen, oxidants, and antioxidants in wound healing - An emerging paradigm. *Alcohol and Wine in Health and Disease* 2002, *957*, 239-249.
14. Liu, X. Y.; Sen, S.; Liu, J. Y.; Kulaots, I.; Geohegan, D.; Kane, A.; Puretzky, A. A.; Rouleau, C. M.; More, K. L.; Palmore, G. T. R.; Hurt, R. H., Antioxidant Deactivation on Graphenic Nanocarbon Surfaces. *Small* 2011, *7* (19), 2775-2785.
15. Pasquini, L. M.; Sekol, R. C.; Taylor, A. D.; Pfefferle, L. D.; Zimmerman, J. B., Realizing Comparable Oxidative and Cytotoxic Potential of Single-and Multiwalled Carbon Nanotubes through Annealing. *Environ. Sci. Technol.* 2013, *47* (15), 8775-8783.
16. Wang, L.; Ambrosi, A.; Pumera, M., "Metal-Free" Catalytic Oxygen Reduction Reaction on Heteroatom-Doped Graphene is Caused by Trace Metal Impurities. *Angew Chem Int Edit* 2013, *52* (51), 13818-13821.
17. Ambrosi, A.; Pumera, M., The CVD graphene transfer procedure introduces metallic impurities which alter the graphene electrochemical properties. *Nanoscale* 2014, *6* (1), 472-476.
18. Guo, L.; Morris, D. G.; Liu, X.; Vaslet, C.; Hurt, R. H.; Kane, A. B., Iron Bioavailability and Redox Activity in Diverse Carbon Nanotube Samples. *Chem. Mater.* 2007, *19* (14), 3472-3478.
19. Teo, W. Z.; Chng, E. L.; Sofer, Z.; Pumera, M., Cytotoxicity of halogenated graphenes. *Nanoscale* 2014, *6* (2), 1173-80.

20. Chng, E. L.; Sofer, Z.; Pumera, M., Cytotoxicity profile of highly hydrogenated graphene. *Chemistry (Easton)* 2014, 20 (21), 6366-73.
21. Chng, E. L. K.; Pumera, M., The Toxicity of Graphene Oxides: Dependence on the Oxidative Methods Used. *Chemistry – A European Journal* 2013, 19 (25), 8227-8235.
22. Lucente-Schultz, R. M.; Moore, V. C.; Leonard, A. D.; Price, B. K.; Kosynkin, D. V.; Lu, M.; Partha, R.; Conyers, J. L.; Tour, J. M., Antioxidant Single-Walled Carbon Nanotubes. *J. Am. Chem. Soc.* 2009, 131 (11), 3934-3941.
23. Chiang, L. Y.; Lu, F. J.; Lin, J. T., Free-Radical Scavenging Activity of Water-Soluble Fullerenols. *J Chem Soc Chem Comm* 1995, (12), 1283-1284.
24. Okuda, K.; Hirota, T.; Hirobe, M.; Nagano, T.; Mochizuki, M.; Mashino, T., Synthesis of various water-soluble C-60 derivatives and their superoxide-quenching activity. *Fullerene Sci. Technol.* 2000, 8 (3), 127-142.
25. Fenoglio, I.; Tomatis, M.; Lison, D.; Muller, J.; Fonseca, A.; Nagy, J. B.; Fubini, B., Reactivity of carbon nanotubes: Free radical generation or scavenging activity? *Free Radical Biol. Med.* 2006, 40 (7), 1227-1233.
26. Galano, A., Carbon nanotubes as free-radical scavengers. *J Phys Chem C* 2008, 112 (24), 8922-8927.
27. Martin, R.; Menchon, C.; Apostolova, N.; Victor, V. M.; Alvaro, M.; Herance, J. R.; Garcia, H., Nano-Jewels in Biology. Gold and Platinum on Diamond Nanoparticles as Antioxidant Systems Against Cellular Oxidative Stress. *ACS nano* 2010, 4 (11), 6957-6965.
28. Bitner, B. R.; Marcano, D. C.; Berlin, J. M.; Fabian, R. H.; Cherian, L.; Culver, J. C.; Dickinson, M. E.; Robertson, C. S.; Pautler, R. G.; Kent, T. A.; Tour, J. M., Antioxidant Carbon Particles Improve Cerebrovascular Dysfunction Following Traumatic Brain Injury. *ACS nano* 2012, 6 (9), 8007-8014.
29. Burton, G. W.; Ingold, K. U., Beta-Carotene - an Unusual Type of Lipid Antioxidant. *Science* 1984, 224 (4649), 569-573.

30. Martinez, A.; Galano, A., Free Radical Scavenging Activity of Ultrashort Single-Walled Carbon Nanotubes with Different Structures through Electron Transfer Reactions. *J Phys Chem C* 2010, *114* (18), 8184-8191.
31. Morita, Y.; Suzuki, S.; Sato, K.; Takui, T., Synthetic organic spin chemistry for structurally well-defined open-shell graphene fragments. *Nat. Chem.* 2011, *3* (3), 197-204.
32. Wright, J. S.; Johnson, E. R.; DiLabio, G. A., Predicting the activity of phenolic antioxidants: Theoretical method, analysis of substituent effects, and application to major families of antioxidants. *J. Am. Chem. Soc.* 2001, *123* (6), 1173-1183.
33. Bors, W.; Heller, W.; Michel, C.; Saran, M., Flavonoids as Antioxidants - Determination of Radical-Scavenging Efficiencies. *Methods Enzymol.* 1990, *186*, 343-355.
34. Fukumoto, L. R.; Mazza, G., Assessing antioxidant and prooxidant activities of phenolic compounds. *J. Agric. Food. Chem.* 2000, *48* (8), 3597-604.
35. Graf, E.; Mahoney, J. R.; Bryant, R. G.; Eaton, J. W., Iron-Catalyzed Hydroxyl Radical Formation - Stringent Requirement for Free Iron Coordination Site. *J. Biol. Chem.* 1984, *259* (6), 3620-3624.
36. Lastoskie, C.; Gubbins, K. E.; Quirke, N., Pore-Size Heterogeneity and the Carbon Slit Pore - a Density-Functional Theory Model. *Langmuir* 1993, *9* (10), 2693-2702.
37. Wang, Z.; von dem Bussche, A.; Kabadi, P. K.; Kane, A. B.; Hurt, R. H., Biological and environmental transformations of copper-based nanomaterials. *ACS nano* 2013, *7* (10), 8715-27.
38. Valentao, P.; Fernandes, E.; Carvalho, F.; Andrade, P. B.; Seabra, R. M.; Bastos, M. L., Antioxidant activity of Centaurium erythraea infusion evidenced by its superoxide radical scavenging and xanthine oxidase inhibitory activity. *J. Agric. Food. Chem.* 2001, *49* (7), 3476-9.
39. Yin, J. J.; Lao, F.; Fu, P. P.; Wamer, W. G.; Zhao, Y.; Wang, P. C.; Qiu, Y.; Sun, B.; Xing, G.; Dong, J.; Liang, X. J.; Chen, C., The scavenging of reactive oxygen species and

the potential for cell protection by functionalized fullerene materials. *Biomaterials* 2009, 30 (4), 611-21.

40. Blois, M. S., Antioxidant Determinations by the Use of a Stable Free Radical. *Nature* 1958, 181 (4617), 1199-1200.

41. Re, R.; Pellegrini, N.; Proteggente, A.; Pannala, A.; Yang, M.; Rice-Evans, C., Antioxidant activity applying an improved ABTS radical cation decolorization assay. *Free Radical Biol. Med.* 1999, 26 (9-10), 1231-1237.

42. Halliwell, B.; Gutteridge, J. M. C., Formation of a Thiobarbituric-Acid-Reactive Substance from Deoxyribose in the Presence of Iron Salts - the Role of Superoxide and Hydroxyl Radicals. *FEBS Lett.* 1981, 128 (2), 347-352.

43. Peyrat-Maillard, M. N.; Cuvelier, M. E.; Berset, C., Antioxidant activity of phenolic compounds in 2,2'-azobis (2-amidinopropane) dihydrochloride (AAPH)-induced oxidation: Synergistic and antagonistic effects. *J. Am. Oil Chem. Soc.* 2003, 80 (10), 1007-1012.

44. Ohkawa, H.; Ohishi, N.; Yagi, K., Reaction of Linoleic-Acid Hydroperoxide with Thiobarbituric Acid. *J. Lipid Res.* 1978, 19 (8), 1053-1057.

45. Litjens, R. A. J.; Quickenden, T. I.; Freeman, C. G., Visible and near-ultraviolet absorption spectrum of liquid water. *Appl. Opt.* 1999, 38 (7), 1216-1223.

46. Berg, J. M.; Romoser, A.; Banerjee, N.; Zebda, R.; Sayes, C. M., The relationship between pH and zeta potential of similar to 30 nm metal oxide nanoparticle suspensions relevant to in vitro toxicological evaluations. *Nanotoxicology* 2009, 3 (4), 276-283.

47. Chen, Y.; Guo, F.; Qiu, Y.; Hu, H.; Kulaots, I.; Walsh, E.; Hurt, R. H., Encapsulation of Particle Ensembles in Graphene Nanosacks as a New Route to Multifunctional Materials. *ACS nano* 2013.

48. Lozano-Castello, D.; Cazorla-Amoros, D.; Linares-Solano, A., Usefulness of CO₂ adsorption at 273 K for the characterization of porous carbons. *Carbon* 2004, 42 (7), 1233-1242.

49. Qiu, Y.; Guo, F.; Hurt, R.; Külaots, I., Explosive thermal reduction of graphene oxide-based materials: Mechanism and safety implications. *Carbon* 2014, 72 (0), 215–223.
50. Guo, F.; Creighton, M.; Chen, Y. T.; Hurt, R.; Kulaots, I., Porous structures in stacked, crumpled and pillared graphene-based 3D materials. *Carbon* 2014, 66, 476-484.
51. Ku, Y.; Tu, Y. H.; Ma, C. M., Effect of hydrogen peroxide on the decomposition of monochlorophenols by sonolysis in aqueous solution. *Water Res.* 2005, 39 (6), 1093-1098.
52. De Visscher, A.; Van Langenhove, H., Sonochemistry of organic compounds in homogeneous aqueous oxidising systems. *Ultrason. Sonochem.* 1998, 5 (3), 87-92.
53. Burkitt, M. J.; Gilbert, B. C., Model Studies of the Iron-Catalyzed Haber-Weiss Cycle and the Ascorbate-Driven Fenton Reaction. *Free Radic. Res. Commun.* 1990, 10 (4-5), 265-280.
54. Nappi, A. J.; Vass, E., Comparative studies of enhanced iron-mediated production of hydroxyl radical by glutathione, cysteine, ascorbic acid, and selected catechols. *Biochim. Biophys. Acta* 1997, 1336 (2), 295-302.
55. Creighton, M. A.; Rangel-Mendez, J. R.; Huang, J.; Kane, A. B.; Hurt, R. H., Graphene-Induced Adsorptive and Optical Artifacts During In Vitro Toxicology Assays. *Small* 2013, 9 (11), 1921-1927.
56. Limon-Pacheco, J.; Gonsbatt, M. E., The role of antioxidants and antioxidant-related enzymes in protective responses to environmentally induced oxidative stress. *Mutat. Res.* 2009, 674 (1-2), 137-47.
57. Nishikimi, M., Oxidation of Ascorbic-Acid with Superoxide Anion Generated by Xanthine Xanthine Oxidase System. *Biochem. Biophys. Res. Commun.* 1975, 63 (2), 463-468.
58. Halliwell, B.; Foyer, C. H., Ascorbic acid, metal ions and the superoxide radical. *Biochem. J* 1976, 155 (3), 697-700.
59. Niki, E., Action of Ascorbic-Acid as a Scavenger of Active and Stable Oxygen Radicals. *Am. J. Clin. Nutr.* 1991, 54 (6), S1119-S1124.

60. Murthy, C. N.; Geckeler, K. E., The water-soluble [small beta]-cyclodextrin-[60]fullerene complex. *Chem. Commun.* 2001, (13), 1194-1195.
61. Xia, X. R.; Monteiro-Riviere, N. A.; Riviere, J. E., Intrinsic biological property of colloidal fullerene nanoparticles (nC(60)): Lack of lethality after high dose exposure to human epidermal and bacterial cells. *Toxicol. Lett.* 2010, 197 (2), 128-134.
62. Valko, M.; Leibfritz, D.; Moncol, J.; Cronin, M. T.; Mazur, M.; Telser, J., Free radicals and antioxidants in normal physiological functions and human disease. *Int. J. Biochem. Cell Biol.* 2007, 39 (1), 44-84.
63. Ballatori, N.; Krance, S. M.; Notenboom, S.; Shi, S.; Tieu, K.; Hammond, C. L., Glutathione dysregulation and the etiology and progression of human diseases. *Biol. Chem.* 2009, 390 (3), 191-214.
64. Zhu, Q. Y.; Zhang, X. M.; Fry, A. J., Bond dissociation energies of antioxidants. *Polym. Degrad. Stab.* 1997, 57 (1), 43-50.
65. Kim, S.; Zhou, S.; Hu, Y. K.; Acik, M.; Chabal, Y. J.; Berger, C.; de Heer, W.; Bongiorno, A.; Riedo, E., Room-temperature metastability of multilayer graphene oxide films. *Nat Mater* 2012, 11 (6), 544-549.
66. Everett, S. A.; Dennis, M. F.; Patel, K. B.; Maddix, S.; Kundu, S. C.; Willson, R. L., Scavenging of nitrogen dioxide, thiyl, and sulfonyl free radicals by the nutritional antioxidant beta-carotene. *J. Biol. Chem.* 1996, 271 (8), 3988-3994.
67. Qin, Y.; Wheeler, R. A., Density-functional-derived structures, spin properties, and vibrations for phenol radical cation. *J. Phys. Chem.* 1996, 100 (25), 10554-10563.
68. Becker, E. M.; Nissen, L. R.; Skibsted, L. H., Antioxidant evaluation protocols: Food quality or health effects. *Eur. Food Res. Technol.* 2004, 219 (6), 561-571.
69. Ramanathan, T.; Abdala, A. A.; Stankovich, S.; Dikin, D. A.; Herrera-Alonso, M.; Piner, R. D.; Adamson, D. H.; Schniepp, H. C.; Chen, X.; Ruoff, R. S.; Nguyen, S. T.; Aksay,

- I. A.; Prud'homme, R. K.; Brinson, L. C., Functionalized graphene sheets for polymer nanocomposites. *Nat Nanotechnol* 2008, 3 (6), 327-331.
70. Lewicka, Z. A.; Benedetto, A. F.; Benoit, D. N.; Yu, W. W.; Fortner, J. D.; Colvin, V. L., The structure, composition, and dimensions of TiO₂ and ZnO nanomaterials in commercial sunscreens. *J. Nanopart. Res.* 2011, 13 (9), 3607-3617.
71. Dunford, R.; Salinaro, A.; Cai, L.; Serpone, N.; Horikoshi, S.; Hidaka, H.; Knowland, J., Chemical oxidation and DNA damage catalysed by inorganic sunscreen ingredients. *FEBS Lett.* 1997, 418 (1-2), 87-90.
72. Xiao, J.; Chen, W.; Wang, F.; Du, J., Polymer/TiO₂ Hybrid Nanoparticles with Highly Effective UV-Screening but Eliminated Photocatalytic Activity. *Macromolecules* 2013, 46 (2), 375-383.
73. Wakefield, G.; Green, M.; Lipscomb, S.; Flutter, B., Modified titania nanomaterials for sunscreen applications - reducing free radical generation and DNA damage. *Mater. Sci. Technol.* 2004, 20 (8), 985-988.
74. Kempaiah, R.; Salgado, S.; Chung, W. L.; Maheshwari, V., Graphene as membrane for encapsulation of yeast cells: protective and electrically conducting. *Chem. Commun.* 2011, 47 (41), 11480-11482.
75. Wen, X.; Garland, C. W.; Hwa, T.; Kardar, M.; Kokufuta, E.; Li, Y.; Orkisz, M.; Tanaka, T., Crumpled and Collapsed Conformations in Graphite Oxide Membranes. *Nature* 1992, 355 (6359), 426-428.
76. Luo, J. Y.; Jang, H. D.; Sun, T.; Xiao, L.; He, Z.; Katsoulidis, A. P.; Kanatzidis, M. G.; Gibson, J. M.; Huang, J. X., Compression and Aggregation-Resistant Particles of Crumpled Soft Sheets. *ACS nano* 2011, 5 (11), 8943-8949.
77. Ma, X. F.; Zachariah, M. R.; Zangmeister, C. D., Crumpled Nanopaper from Graphene Oxide. *Nano Lett.* 2012, 12 (1), 486-489.

78. Linsebigler, A. L.; Lu, G. Q.; Yates, J. T., Photocatalysis on TiO_2 Surfaces - Principles, Mechanisms, and Selected Results. *Chem. Rev.* 1995, 95 (3), 735-758.
79. Ghiazza, M.; Tomatis, M.; Doublier, S.; Grendene, F.; Gazzano, E.; Ghigo, D.; Fubini, B., Carbon in intimate contact with quartz reduces the biological activity of crystalline silica dusts. *Chem. Res. Toxicol.* 2013, 26 (1), 46-54.
80. Sellappan, R.; Sun, J.; Galeckas, A.; Lindvall, N.; Yurgens, A.; Kuznetsov, A. Y.; Chakarov, D., Influence of graphene synthesizing techniques on the photocatalytic performance of graphene- TiO_2 nanocomposites. *Phys. Chem. Chem. Phys.* 2013, 15 (37), 15528-37.
81. Zhang, Y.; Tang, Z. R.; Fu, X.; Xu, Y. J., TiO_2 -graphene nanocomposites for gas-phase photocatalytic degradation of volatile aromatic pollutant: is TiO_2 -graphene truly different from other TiO_2 -carbon composite materials? *ACS nano* 2010, 4 (12), 7303-14.

**CHAPTER 5 GO THERMAL AEROSOL
PROCESSING AS A VERSATILE WAY
TO BUILD 3D ASSEMBLIES**

This chapter is based on published work by Y. Chen, F. Guo, Y. Qiu, H. Hu, I. Kulaots, E. Walsh and R. Hurt *ACS Nano*, 2013

5.1 Introduction

GO in aqueous solution is a relatively flexible material and can easily fold/unfold under applied force. This particular feature makes it possible to use water surface tension to manipulate two dimension (2D) GO sheets into various dimensions and structures of interests. One instance is the self assembling of GO Langmuir-Blodgett film to construct 2D structure –monolayer assembly using water surface tension.¹ However, it is more challenging to build well-controlled 3D structures, as it would require the 3D manipulation of water droplets. One effective way to achieve this is aerosol processing,²⁻⁴ which is a two-step process. The first step is the transport of dispersed GO sheets in the bulk solution onto the air-water interface of droplet and the second step involves water droplet-actuated scrolling, folding and collapsing. The first step is thermodynamically favorable due to the drop of free energy as the solid-liquid interface and the liquid-gas interface are destructed and an equal area solid-gas interface is created. The second step involves a “guide and glide” assembly process in which the water droplet drags the GO sheets inward during vaporization and guide the formation of crumpled and plastically deformed structures with kinks.² This crumpled structure has been shown to be a versatile platform for building 3D graphene composite assemblies with various cargo materials (nanoparticles, polymers or salts) being encapsulated.²⁻⁴ Here using aerosol processing as an effective and relatively simple way to construct multifunctional composite materials is explored.

The crumpled structure can be synthesized through aerosol processing at room temperature if the partial pressure of water in the drying chamber is enough low. This can

be achieved through replacing tube furnace with diffusion dryer.² However, the unreduced crumpled GO product can spontaneously unfold rapidly if dispersed in aqueous solution due to the highly hygroscopic nature of GO, which attracts water molecules to penetrate into the interlayer space of stacked sheets of the crumpled GO shell, expand the interlayer space and unfold the 3D assembly into smaller 2D flakes. Thermal treatment is thus necessary to stabilize the crumpled structure in such a system. Once the pristine GO changes into rGO under heating the strong Van der Waals interaction between rGO layers firmly holds the sheets together, thus prevents the introduction of water molecules and stabilizes the 3D structure when dispersed in water. In addition to reducing GO the thermal treatment could also play other roles. In this chapter, different systems will be investigated and mechanisms will be discussed.

5.2 Materials and methods

5.2.1 Materials

SP-1 graphite powder was purchased from Bay Carbon (Bay City, MI). $K_2S_2O_8$, $KMnO_4$, $FeCl_3 \cdot 6H_2O$, $HAuCl_4$, Fe_3O_4 and CuO NPs with a particle size of <50 nm and Acetone were purchased from Sigma Aldrich Chemicals Co (St. Louis, MO). 20 nm Fe_3O_4 and 50 nm $BaTiO_3$ NPs were purchased from U.S. Research Nanomaterials Inc (Houston, TX). $NaNO_3$, P_2O_5 , HCl and concentrated H_2SO_4 (98 %) were purchased from Fisher Scientific Inc. (Pittsburgh, PA). GO was synthesized with the modified Hummers method with pre-oxidation and two-step acid-acetone wash as described in previous chapters.

5.2.2 Methods

5.2.2.1 Synthesis of Au NPs

40 nm Au NPs were synthesized by the reduction of 50 mL of HAuCl_4 in the presence of citrate and tannic acids. Briefly, 24 mL 0.25 % (w/v) HAuCl_4 was added into 115 mL nanopure water and heated up to 60 °C. Under vigorous stirring, a solution mixed by 5 mL 1 % (w/v) tannic acid, 0.2 mL 1 % (w/v) trisodium citrate and 5 mL 2.5 mM K_2CO_3 was rapidly added. The color changed rapidly after the addition of the reducing agents. The solution was then heated to boiling for 10-15 min and cooled down to room temperature. After vigorous washing with nanopure water and concentration, the obtained stock solution of 10 mg/mL can be stored in refrigerator for weeks.

5.2.2.2 Synthesis of FeOOH Nanorods:

FeOOH nanorods with average length around 200 nm were synthesized by modified hydrothermal methods reported before.⁵ $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$ was dissolved in 100 mL deionized water to prepare 0.02 M FeCl_3 solution, and then heated at 80°C with mechanical stirring in water bath for 12 hr to obtain FeOOH nanorods. These particles were collected by centrifugation and washed with DI water several times.

5.2.2.3 Aerosol processing of GO

Colloidal suspensions of GO (0.5 mg/mL) and one or more cargo nanoparticles were subject to sonication together to obtain a dispersion and then ultrasonically nebulized to aerosol microdroplets (nebulizing feed rate: 10 mL/h) suspended in nitrogen gas (0.7 L/min) and passed directly into a horizontal benchtop electric tube furnace operated at desired temperature as explained in Chen et al.² The products, which are cargo-filled graphene

nanosacks, were collected on a heated (~90 °C) Teflon filter (PTU024750, Sterlitech Co.) or onto a small silicon wafer chip (5 x 5 mm, Ted Pella, Inc.) or copper grid (Ted Pella, Inc.) placed on top for SEM or TEM analysis. The controlled release experiments used nebulizing feed suspensions containing GO, CsCl, and the water-soluble polymer sodium carboxymethylcellulose (Sigma Aldrich) at a mass ratio CMC/GO/CsCl of 0.5:1:4. The primary drying temperature was reduced to 80-100 °C and followed by post- carbonization of CMC at 500 °C by slow heating in a batch tube furnace for 30 min.

5.2.2.4 Characterization

SEM (LEO 1530 VP at 15 kV), TEM (Philips CM 20 at 200 kV), and HRTEM (JEOL JEM-2010 at 200 kV) were carried out at the Brown IMNI Electron Microscopy Facility. Zeta-potential and particle size distributions were measured in DI water using a Malvern Nano-ZS dynamic light scattering (DLS) instrument. Before measuring the zeta-potential, the pH value of the suspension was measured by using an Orion 5-Star Plus pH/ORP/ISE/conductivity/ DO meter (Thermo Scientific Inc.). Prior to zeta-potential measurement, the pH values of nanoparticle suspensions were adjusted with 0.1 M NaOH or 0.1 M HCl to match the pH in the binary suspensions used in the nebulizer feed. A Quantachrome Auto- sorb-1 instrument was used to measure N₂ vapor isotherms at 77 K, from which Brunauer_Emmett_Teller (BET) surface areas and pore size distributions applying nonlocal density functional theory (DFT) with a carbon slit pore model³⁴ were calculated.

5.2.2.5 MRI/CT/Magnetometry

A dry sample of the hybrid Au/Fe₃O₄- loaded graphene nanosacks was assessed for developed (saturation) magnetization and hysteresis (retentivity and coercivity) using a vibrating sample magnetometer (Lakeshore VSM model 7300). A 15 mg sample was placed

in a 5 mm plastic capsule with a PTFE plug to constrain the material. The applied magnetic field strength was swept from -0.8 to 0.8 T while 441 measurements were taken. Three versions of loaded nanosacks were assessed for imaging properties: iron oxide nanoparticles (Fe_3O_4) as an MRI agent, gold nanoparticles as an X-ray CT agent, and a hybrid Au/ Fe_3O_4 version for multimodal X-ray and MRI application. The hybrid sample was the same one used for the magnetometry assessment. Iron oxide is used in clinically approved MRI T_2 agents and produced a strong susceptibility effect. Gold is being investigated for use in nanoparticle contrast and therapy agents owing to its strong X-ray attenuation and scattering properties. The mass attenuation coefficient of gold at 100 keV is $5.16 \text{ cm}^2/\text{g}$, which is 93% that of lead for the same photon energy.⁶⁻⁷

For MRI, nanosacks filled with Fe_3O_4 or Au/ Fe_3O_4 nanoparticles were suspended in CMC gel (1 %) at concentrations of 0.5, 1, 2, 5, 10, 20, and 50 $\mu\text{g}/\text{mL}$ for Fe_3O_4 and 0.05, 0.1, 0.5, 1, 5, 10, 50, 100, 500, and 2000 $\mu\text{g}/\text{mL}$ for the hybrid. These preparations were placed in 5 mL vials along with control preparations of the same concentrations but not containing graphene nanosacks. In addition, control samples were included in the form of distilled water and gel with no added material. MRI data were acquired at the Brown University Magnetic Resonance Facility using a 3 T Siemens Tim Trio scanner. Samples were placed in a 32 channel receive resonator, and the volume body resonator was used for transmit. Cross section tomographic images were obtained using a spin echo acquisition for mapping T_2 .

T_1 -weighted images were obtained using an inversion recovery contrast preparation prior to the spin echo readout. An inversion recovery series was used for mapping T_1 . Slice thickness was 3 mm, and first-order shimming was performed to optimize static magnetic field homogeneity over the samples. In-plane resolution was 0.7 mm using a 256×256 reconstruction matrix. Repetition time for the spin echo acquisition was 2000 ms and 4000

ms for the inversion recovery series. Echo times for the spin echo acquisition ranged from 10 to 240 ms in 10 ms steps (24 echoes). For the inversion recovery series, inversion delay times were 100, 200, 300, 400, 600, 1000, and 2000 ms. Relaxation time constant maps (T_1 , T_2) were computed on a pixel basis using three-parameter least-squares fits to the signal equations:

$$S(TE) = S_0 e^{-\frac{TE}{T_2}} \quad (5.1)$$

$$S(TI) = S_0 (1 - 2e^{-\frac{TI}{T_1}} - 2e^{-\frac{TR}{T_1}}) \quad (5.2)$$

where S_0 is the equilibrium (maximum possible) magnetization signal, TE is the echo time (for the spin and gradient echo acquisitions), TR is the repetition time, and TI is the inversion time. Relaxation rate maps were taken as the reciprocals of the relaxation time constant maps (e.g., $R_2 = 1/T_2$) for computation of relaxivity (change in relaxation rate per unit change in concentration of agent).

For X-ray CT studies, the same G/Au and G/Au/Fe₃O₄ samples were used as for MRI. For the G/Au sample, concentrations of 20, 40, 200, 400, and 2000 µg/mL were used. A General Electric Light Speed VCT scanner (Department of Diagnostic Imaging, Rhode Island Hospital) was used for acquisition of tomographic cross section images, providing the same view as produced by the MRI scans. Slice thickness was 3 mm with a reconstruction matrix of 512 x 512 and an in-plane resolution of 0.5 mm. The X-ray tube current was 200 mAs. Images were acquired for X-ray tube voltages of 80, 100, 120, and 140 kVp. Contrast was determined as the difference in CT number (Hounsfield units) between the control gel sample and the samples containing contrast material. CT numbers were taken as the means of circular regions of interest from the center 80% of the cross sections. Hounsfield units are related to X-ray attenuation coefficient by

$$CT(x, y) = 1000 \left(\frac{\mu(x, y) - \mu_{H_2O}}{\mu_{H_2O}} \right) \quad (5.3)$$

where μ is the attenuation coefficient and μ_{H_2O} is the attenuation coefficient for water. The Hounsfield unit scale is normalized such that the attenuation of air corresponds to a CT number of -1000; the CT number of water is 0, with dense bone having a CT number of approximately 3000. In X-ray CT images, the pixel intensities are the CT numbers corresponding to the pixel locations.

5.3 Results and discussions

5.3.1 Thermal aerosol synthesis of crumpled graphene nanosacks

Figure 5.1 shows the aerosol encapsulation process and example structures made from binary feed solutions, meaning those containing graphene oxide (GO) and one cargo particle type. Here an aqueous dispersion containing the cargo and graphene oxide is ultrasonically nebulized, and the microdroplet mist is carried by an inert gas flow through an electrically heated furnace. The dried nanosacks are collected on a polytetrafluorethylene filter (Figure 5.1A), and in the most common experiment, the furnace temperature is set at 500-600 °C, converting the GO to reduced graphene oxide (rGO). We observed successful encapsulation with many of these binary systems, but not all. Some systems formed gels or visible precipitates in the feed suspension that led to nonuniform products or significant numbers of isolated unwrapped nanoparticles (e.g., in the case of CuO NPs and Co NPs functionalized with L-cysteine ethyl ester). Other systems led to good encapsulation even though the nanoparticle cargoes did not form stable colloidal suspensions on their own in water (e.g., Fe₃O₄, BaTiO₃). There are also subtle differences in structure among the successful cases, with some completely encapsulated with a high degree of sack-cargo segregation (Figure

5.1B), while others show particles that appear to be on the outer surface (arrows, Figure 5.1D,E). HRTEM on these systems reveals that some of the nanoparticles are in fact covered by very thin reduced graphene oxide films with a width of ~ 2 nm and are thus “inside” but in the near-surface region. On the basis of these facts, we hypothesized that colloidal interactions involving the cargo particles and the graphene oxide sheets play an important role in successful aerosol encapsulation and the structure of the hybrid material products.

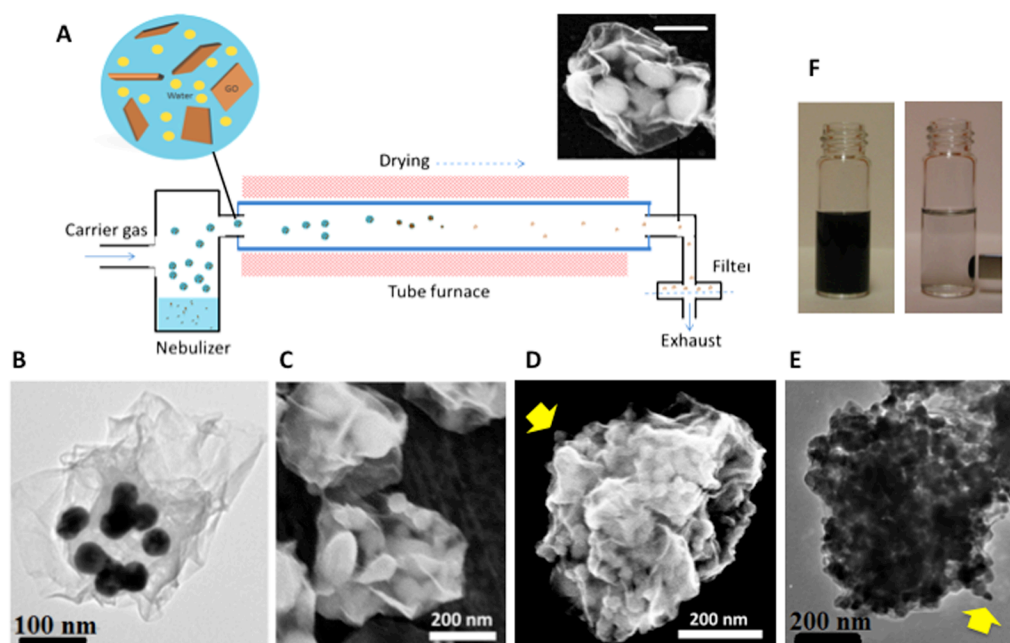


Figure 5.1 Aerosol-phase synthesis of cargo-filled nanosacks and examples of binary materials with a single homogeneous cargo.

(A) The continuous aerosol fabrication process leading to Ag-filled nanosacks.² (B-E) electron micrographs of selected binary composite product structures (graphene oxide with single cargo material). (B) tannic-acid-capped AuNPs, (C) Si-NPs, (D)-(E) Fe₃O₄ NPs, (F) the Fe₃O₄ filled nanosacks can be easily cleared from ethanol solution with a hand-held magnet demonstrating stable encapsulation and the absence of free graphene. The particles alone cannot be dispersed in ethanol, so the graphene serves as both a dispersing and encapsulation agent in this case. Arrows on D, E show a minority of particles on or very near the outside surface of the Fe₃O₄-filled nanosacks.

Chen et al proposed a mechanism for sack_cargo formation, in which graphene oxide shows a thermodynamic preference to localize at the air/water interface and its large hydrodynamic size leads to slow diffusion away from the drying front. GO thus collects as a

multilayer film at the droplet outer surface and collapses by capillary forces in the late stages of drying. The NP cargoes in contrast diffuse away from the drying front to localize preferentially in the interior.² This theory was developed for citrate-capped Ag-NPs, which show a large negative surface charge in common with GO, and the original theory was specific for such mutually repelling binary colloids. Some NPs of interest here in the larger sample set, however, have positive charge or are nearly isoelectric, and a more general theory is needed to understand the range of applicability of the aerosol technique and to explain the observations above.

Figure 5.2 shows the elements of a new, general theory of sack-cargo assembly that applies in different electrostatic regimes. The theory will be formulated for binary colloids, but is easily extended to systems with three or more colloidal components. Figure 5.2A shows pH-dependent zeta potentials for a generalized binary colloid, which give rise to three regimes: two electrostatically repulsive regimes at high and low pH where colloidal stability and sack-cargo segregation are expected, and an attractive regime at intermediate pH, where complex association phenomena can occur between the colloidal particle components. To apply this to sack assembly, we measured zeta potentials for all candidate cargo particles in deionized water, but the results were not directly relevant due to differences in pH between the single-component suspensions and the binary suspensions containing GO. We therefore measured the pH of the GO/cargo nebulizing suspensions and measured zeta potentials for GO and particles at the relevant mixture pH (Figure 5.2B). For GO itself, the net surface charge is always negative between pH 2 and 12, and the potential/pH profile has a flat valley above pH 5, which is consistent with carboxylate groups as the primary source of acidity and thus surface charge. Gold NPs were characterized as an example cargo, and are also persistently negative due to their tannic acid capping. The GO/Au system shows only one of the three possible regimes, and its highly repulsive nature leads to good sack-cargo segregation. Iron oxide NPs show a zeta

potential with a strong pH dependence and an isoelectric point near 7.⁸⁻⁹ The GO/Fe₃O₄ system thus shows two of the three possible regimes in Figure 5.2A. The pH of our GO/Fe₃O₄ suspensions varies with GO/Fe₃O₄ ratio, and was measured as pH 3 for the case shown in Figure 5.1. At this pH, the Fe₃O₄ NPs have strongly positive surface charge and must associate with the negatively charged GO. DLVO theory would predict no repulsive barrier to association (heteroaggregation) and indeed we see a single peak in zeta potential measurement in GO/Fe₃O₄ suspensions. We also see a single zeta potential peak for a variety of binary suspensions containing GO and NPs with positive surface charge at the mixture pH. We believe this association explains the ability to disperse and encapsulate Fe₃O₄, which is colloiddally unstable in the absence of GO. In general, the association of NPs with GO can aid in NPs dispersion, but may lead to charge neutralization, GO-GO bridging and aggregation. Small aggregates may be incorporated into the 5 μ m droplets formed by ultrasound, but larger aggregates or gel networks cannot be uniformly nebulized into microdroplets leading to poor encapsulation performance as seen for CuO and Co under some conditions. Some metal and metal oxide NPs are also unstable to dissolution, including Ag,¹⁰ ZnO¹¹ and CuO.¹² Here we observed the zeta potential of CuO/GO mixtures to be higher than either CuO or GO alone, and to form gel phases, which we believe is due to CuO partial dissolution and Cu²⁺ binding to GO-COO⁻ groups that neutralize surface charge and destroy colloidal stability. We found that NaOH titration of GO/CuO-NP suspension from pH 5.3 to 10 moves the system from the attractive regime to the repulsive regime and suppresses dissolution, allowing successful CuO encapsulation. Titration to basic pH appears to be useful to avoid metal and metal oxide dissolution during encapsulation.

Figure 5.2C shows the range of NPs used in this study and a conceptual model of the assembly process in the attractive and repulsive regimes. The repulsive regime leads to separate GO and nanoparticle phases, whose differing diffusion rates lead to good

sack-cargo separation during drying (Figure 5.1B). The attractive regime in contrast leads to GO-particle attachment, which reduces the degree of sack-cargo segregation, and leads to the presence of some particles on the external surface or in the near-surface regime intercalated between graphene sheets as seen in Figure 5.1D, E. This conceptual model explains why a wide variety of colloidal particles can be encapsulated by the aerosol technique, including those that are colloidal unstable on their own. Another useful result of this model is that when colloidal instability and/or poor encapsulation is observed, base titration to high pH is an effective technique to establish the repulsive regime where GO and the cargo particle both carry negative surface charges and form stable binary suspensions. A further benefit of base titration is the suppression of metal or metal oxide dissolution, which is typically proton mediated.¹⁰

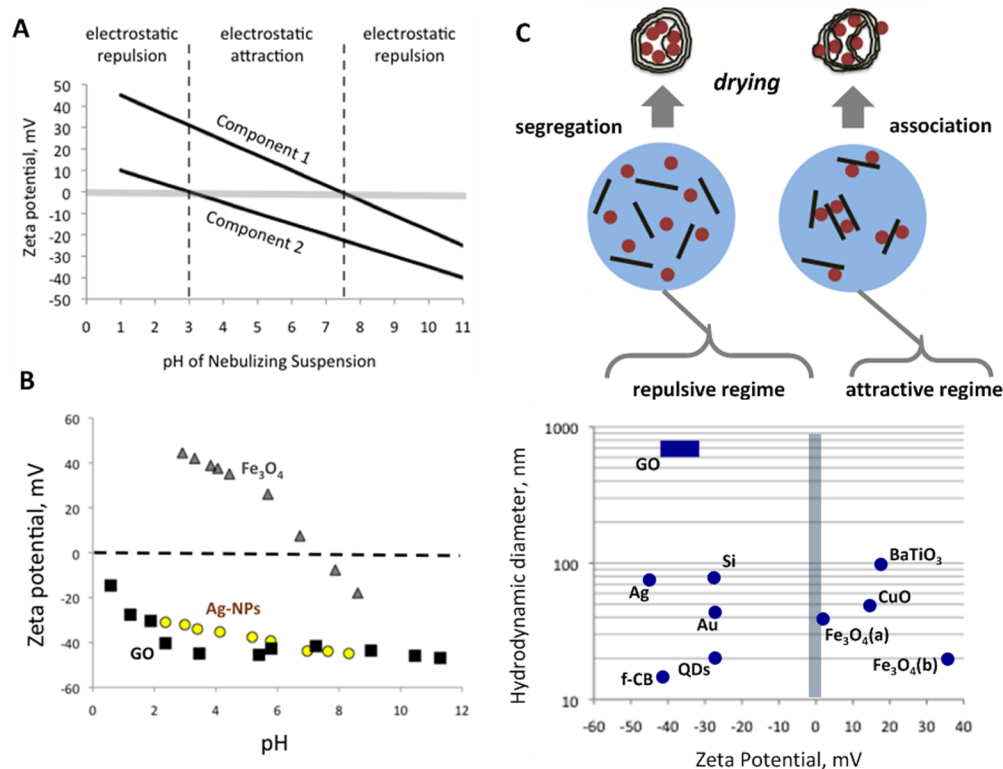


Figure 5.2 Proposed theory for colloidal sack-cargo assembly in binary systems.

(A) pH-dependent zones of electrostatic attraction and repulsion (general behavior). (B) Specific behaviors of graphene oxide and two example cargo materials. The acidic nature of GO gives it a negative colloidal surface charge throughout the pH range 2-12. Only one repulsive regime is seen for the G/Au system. The G/Fe₃O₄ system shows one attractive regime (pH < 7) and one repulsive regime (pH > 7). (C) Map of size and surface charge for sack-cargo systems studied here, and proposed assembly mechanisms in the repulsive and attractive regimes. The hydrodynamic size of all cargoes is smaller than that of GO, allowing fast diffusion of the cargo away from the drying droplet front and thus diffusion segregation and sack-cargo separation in the colloidal repulsive regime where the particles act independently of the GO sheets.

5.3.2 Nanosack sealing and controlled release

A second fundamental issue to resolve in nanosack technology relates to nanosack sealing and internal solute release. If GO nanosacks are hermetically sealed, this would be useful for biological application of acutely toxic cargos, which can be isolated from tissue by the GO nanosack acting as a barrier. If however the sacks are open to small molecule transport, then applications as catalyst supports, electrode materials, drug delivery vehicles, and

theranostics are possible. Several independent assays were devised to characterize nanosack sealing and solute exchange. First we studied nitrogen vapor adsorption on nanosacks filled with Fe_3O_4 -NPs as a probe for the accessibility of internal pores and surfaces. Figure 5.3B shows the pore size distribution of Fe_3O_4 -filled GO nanosacks obtained from nitrogen vapor adsorption data analyzed by nonlocal density functional theory (DFT) applying carbon slit pore model.¹³ There is a characteristic mesopore size of about 4 nm, which is similar to that seen in the empty sacks, and possible attributed to the inner space in loop-shaped ridges seen by SEM and TEM.² The Fe_3O_4 -filled graphene nanosacks have a total accessible surface area of 78 m^2/g and about 0.5 cm^3/g of total pore volume (Figure 5.3A). Using a true density of 1.8 g/cm^3 for reduced GO, the porosity, $V_{\text{pores}}/V_{\text{total}}$, is estimated to be 0.66, which suggests the internal spaces are open to the N_2 adsorbate. The surface area and pore volume of the Fe_3O_4 -filled nanosacks lie between those of the empty sacks and the Fe_3O_4 nanoparticles alone as a bulk powder, and are roughly consistent with simple additive relationship for the relevant Fe_3O_4 /graphene mass ratio, which is 5.7:1.

This significant pore volume suggests the sacks are open, though it is not clear that the pores necessarily connect the particle exterior to the internal cargo. Further evidence that sacks are open structures comes from acid dissolution studies. Immersing the Fe_3O_4 -filled sacks in 1 M HCl is seen to etch away the internal particles leaving the sack intact. Further, cesium chloride was used as model solute to study release from the sack interior. We found that exposing CsCl-filled GO nanosacks to humid room air prior to SEM analysis led to water uptake and the release of salts clearly seen in Figure 5.3C. The highly hygroscopic nature of these particles is consistent with the exposure of nanoscale particles of highly soluble CsCl with high surface area to ambient water vapor. In Figure 5.3D, we use a chloride-specific electrode to measure release kinetics. Release from the as-produced nanosacks is too fast to measure with this technique (< 10 sec). The rapid release in the as-produced sacks is

consistent with pore transport over these length scales. With the relevant particle length scale of 100 nm, a porosity values of 0.66 and a Cl^- aqueous diffusion coefficient of $2 \times 10^{-5} \text{ cm}^2/\text{s}$,¹⁴ characteristic diffusion times, L^2/D , are predicted to be on order of 7 μsec if the pores connect the cargo space with the exterior. Each of these tests indicates the sacks are open and capable of quickly releasing small molecule cargoes.

Delivery of theranostic applications will require controlled release behavior rather than the rapid release observed for as-produced sacks. As a proof-of-principle, we added carboxymethyl cellulose (CMC) to see if an additional filler material could provide a diffusion barrier to achieve slow release of the CsCl salt probe. Addition of CMC to the nebulizing suspension led to GO/CsCl/CMC nanosacks, and the 500 °C drying temperature decomposes CMC to volatile matter and amorphous carbon as a pore blocker. Figure 5.3D shows that a portion of the salt release now occurs over much longer time scales, on the order of days. The ability to add water-soluble solutes and fillers to GO nanosacks opens up the possibility for controlled release applications either alone or in combination with diagnostic functions.

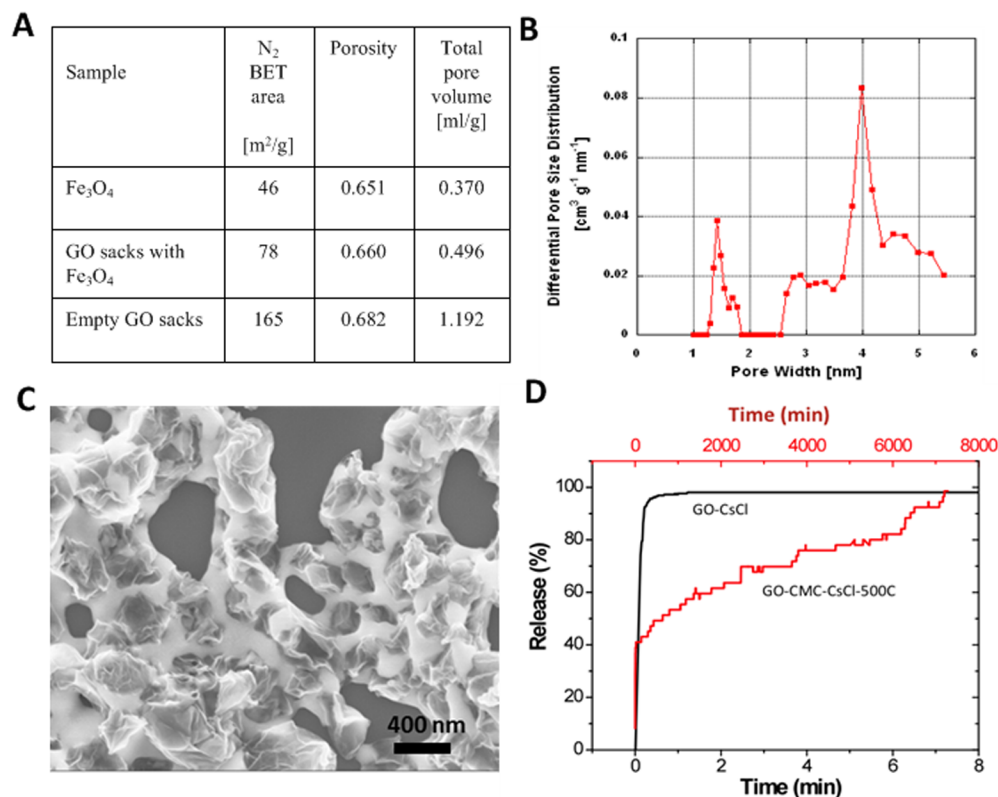


Figure 5.3 Information on graphene nanosack sealing and solute release as relevant to delivery functions.

(A) Brunauer Emmett Teller (BET) surface area, porosity and pore volume of Fe₃O₄, empty nanosacks and Fe₃O₄-filled nanosacks. (B) Pore size distribution of Fe₃O₄-filled nanosacks obtained from nitrogen vapor adsorption isotherms by applying nonlocal density functional theory (DFT) carbon slit pore model. (C) SEM of CsCl-filled GO nanosacks after brief exposure to humid air. Salt-filled sacks (GO:CsCl mass ratio 1:4) are highly hygroscopic and take up atmospheric moisture and release CsCl to form visible films surrounding the sacks. (D) Chloride release rate in DI water for CsCl filled nanosacks with and without carboxymethylcellulose (CMC) as a sealing additive. CMC addition to the aerosol process reduces the dissolution rate of soluble CsCl.

5.3.3 Multimodal imaging

The theory for binary colloids presented in Figure 5.2 is easily extended to systems with $N_{\text{components}} > 2$, which is the regime of interest for multifunctional materials. We chose to demonstrate the multicomponent capability by fabricating dual-purpose magnetically responsive contrast agents for both MRI and X-ray CT. MRI and CT are two of most widely used techniques in clinical diagnostics, and can be applied alone or in combination. Due to

its good availability and relatively low costs, CT is often the preferred technique for pretreatment evaluation of cancer, and offers high patient throughput and high-resolution imaging.¹⁵ MRI is a flexible technique that identifies pathology based upon their difference on T_1 , T_2 (or T_2^*) relaxation time and proton density between diseased and normal tissue. Figure 5.4A-4C shows three different multicomponent graphene sacks of interest in dual CT/MRI imaging, based on G/Au/Fe₃O₄ or G/BaTiO₃/Fe₃O₄ chemistries. Each of the materials was fabricated with a component mass ratio of 1:2:2. The multicomponent nature of these hybrid is most easily seen in Figure 5.4B, in which the lower electron density and small size of the Fe₃O₄ NPs distinguish them from Au, and in Figure 5.4C where the large size of BaTiO₃ NPs distinguishes them from the Fe₃O₄-NPs. High-resolution imaging shows the GO sack is a multilayer structure that conforms to particles in the peripheral region and bridges open spaces between neighboring particles (Figure 5.4D).

The magnetic properties of one set of GO/Au/Fe₃O₄ nanosacks were characterized by vibrating sample magnetometry on 15 mg dry powder samples. We observe a small degree of hysteresis, which is consistent with the 50nm size of the iron oxide nanoparticles in these sacks, and the remanence and coercivity were determined to be 1.040 emu/g and 0.01416 T, respectively. The saturation magnetization was not reached at the maximum applied field of 0.8T, but the maximum developed magnetization noted was 4.52 emu/g at 0.8T. This developed magnetization is substantially larger than that produced by ferritin cage-based iron and manganese contrast agents by a factor of 11.3 and 2.26 respectively.¹⁶⁻¹⁷ Use of single domain superparamagnetic particles as the cargo (8-10 nm) may result in a larger developed magnetization, smaller hysteresis, and even greater T_2 contrast effect.

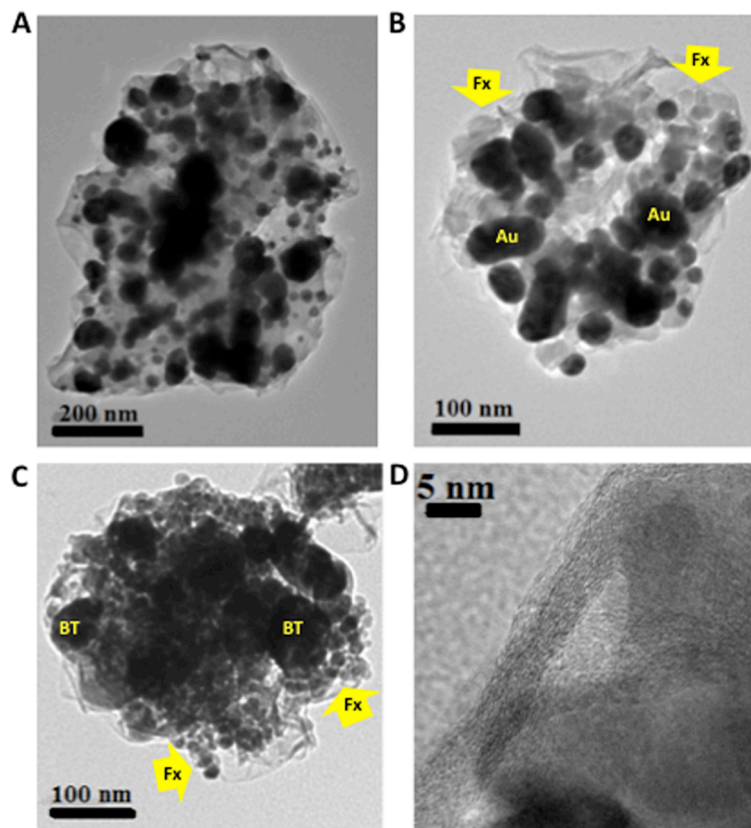


Figure 5.4 Example multifunctional probes as dual CT/MRI contrast agents.

(A) G/Au/Fe₃O₄ hybrid used in the magnetometry and MRI/CT characterization in Figure 5.5. (B) Second G/Au/Fe₃O₄ hybrid with 20 nm Fe₃O₄ NPs (“Fx”) whose small size and low electron density make them visually distinct from the nominal 40 nm Au. (C) G/BaTiO₃/Fe₃O₄ hybrid for GI tract imaging. (D) High resolution image of a multilayer graphene sheet bridging two cargo particles at the outer edge of a G/Fe₃O₄ structure.

Figure 5.5 shows the MRI and CT contrast performances of the GO/Au, GO/Fe₃O₄ and three-way GO/Au/Fe₃O₄ hybrids. The filled graphene nanosacks were suspended in carboxymethylcellulose (CMC) gel in cylindrical 5 ml tubes at a series of increasing concentrations and placed horizontally in the MRI and CT test zones for cross-sectional imaging relative to gel-only controls. Figure 5.5A, B show the MRI behavior of the GO/Fe₃O₄ nanosacks, in the form of T₂ weighted images with an echo time of 30 ms along with a T₂ map computed using the monoexponential fit to the 24-echo data. The Fe₃O₄-filled nanosacks show a strong T₂ contrast effect at ppm doses. From a gel control mean T₂ of 535

ms, T_2 monotonically decreased with increasing concentration reaching a value of 19.1 ms at the concentration of 50 $\mu\text{g/ml}$. The relaxation rate ($R_2 = 1/T_2$) is linear with dose, as expected. Computing relaxivity as the slope of this line gives $r_2=1012 \text{ s}^{-1} (\text{mg/ml})^{-1}$. There was also a weak T_1 effect with an estimated relaxivity $r_1=4.95 \text{ s}^{-1} (\text{mg/ml})^{-1}$. This behavior is expected, as iron oxide based contrast agents are used primarily to reduce T_2 and T_2^* , although some T_1 effect has been described previously.¹⁸ The 3-way GO/Au/ Fe_3O_4 hybrid shows similar behavior (Figure 5.5C) but with reduced relaxivity ($r_2=114.0 \text{ s}^{-1} (\text{mg/ml})^{-1}$) relative to the pure Fe_3O_4 cargo. The hybrid's effect on T_1 was practically negligible with a reduction in T_1 of 9.5% between the gel control at a concentration of 500 $\mu\text{g/ml}$.

Figure 5.5D, E show the X-ray CT contrast performance for the G/Au hybrids, Au-particle controls and gel-only controls. The maximum attenuation was noted at 80 kVp with a mean CT number increase of 57.8 for the 2000 $\mu\text{g/ml}$ Au/nanosack sample. The same concentration for free gold (no nanosacks) produced at mean CT number change of 102, which is consistent with the increased mass fraction of Au in this material. The expected monotonic decrease in attenuation with increasing kVp was interrupted at 120 kVp. This is probably a consequence of the K-edge absorption of gold at 80.725 keV. With an X-ray tube voltage bias of 120 kVp there was a sufficiently large number of photons in the region of 80 keV to allow observation of K-edge attenuation. As expected, a reduction in the quantity of gold in the GO nanosacks reduced the observed attenuation, with a mean CT number change of 25.6 at a concentration of 2000 $\mu\text{g/ml}$ at 80 kVp (Figure 5.5E). Again, there is an increase in attenuation going from 100 kVp to 120 kVp, again indicating the possibility of the K-edge attenuation effect.

The imaging results demonstrate the feasibility of cargo-filled GO nanosacks as contrast agents applicable to MRI and X-ray CT, and the potential for development of versions

suitable for use with both modalities. Note that the degree of T_2 effect is believed to be influenced by water penetration in and out of the nanosack, as is the case for modified ferritin complexes.¹⁶⁻¹⁷ The further development of this application will require optimization of cargoes (single domain magnetic particles), nanosack size (through concentration and droplet size optimization), and functionalization and targeting of the nanosack exterior through well-known coupling chemistries for graphene and related carbon materials.¹⁹⁻²² Further applications may be in spatial frequency heterodyne imaging,²³ which offers promise for use of X-ray scattering in the imaging of object edges with abrupt changes in phase or absorption with low concentrations of the absorbing agent such as gold.

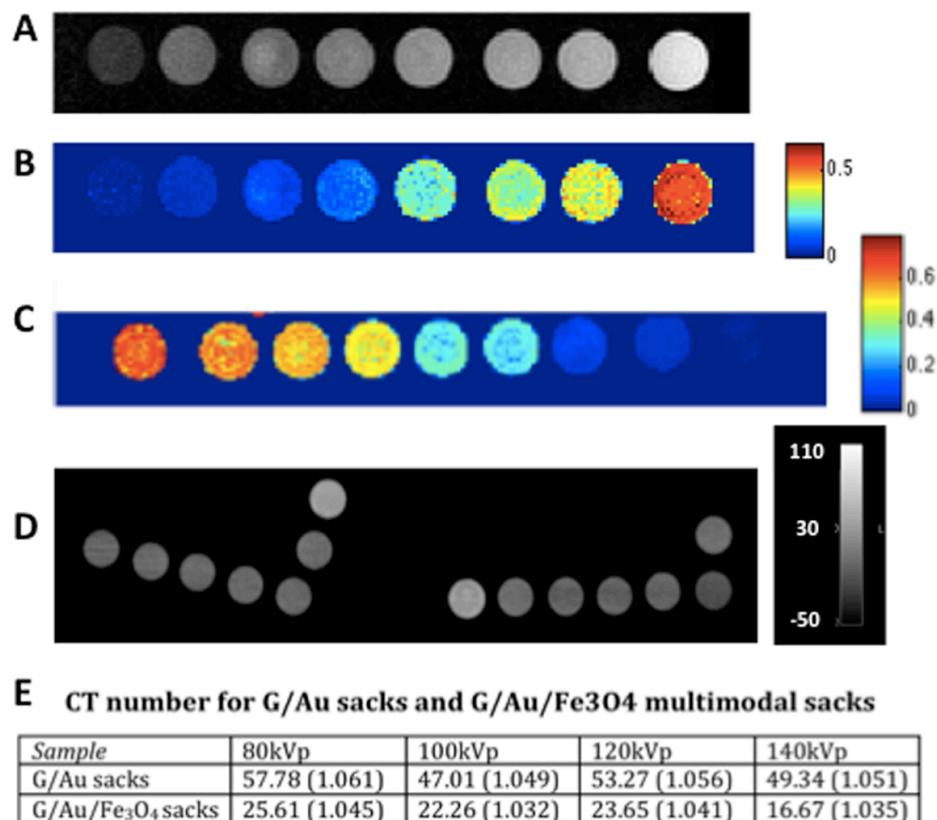


Figure 5.5 Clinical MRI and X-ray CT results showing the contrast performance of G/Fe₃O₄, G/Au and G/Au/Fe₃O₄ multifunctional probes.

(A) T₂ weighted image (TE=30ms) of G/Fe₃O₄ nanosacks in CMC gel. Control (gel alone) is on the right, with nanosack concentration increasing right to left: 0.5, 1, 2, 5, 10, 20, 50 ug/ml. (B) T₂ map computed from the 24 echo image series. Color bar is T₂ in seconds. (C) MRI T₂ map of the G/Au/Fe₃O₄ hybrid. Sack concentrations increase from left to right as 0.05, 0.1, 0.5, 1, 5, 10, 50, 100 and 500 ug/ml. (D) X-ray CT image (80kVp) of the G/Au nanosacks (right-hand L-shaped sequence) and free Au controls (left-hand L-shaped sequence). For the G/Au nanosacks, the sack concentrations in ug/ml from left to right are: 2000, 400, 200, 40, 20, gel control, H₂O control. For the free Au, the particle concentrations are (left to right). 400, 200, 40, 20, gel control with the 2000 ug/ml sample at the top, and a 200 µg/ml sample of empty sacks immediately beneath it. (E) Table of CT results shown as change in CT number with the attenuation (relative to control gel sample) shown in parenthesis.

5.3.4 Carbothermal reduction as a new route for metal/metal oxides production

It is well known that carbon becomes a reducing agent for metal oxides and sulfides at elevated temperature. Ellingham diagram plots the standard free energy of formation of metal oxides/sulfides and carbon oxides against temperature. The standard free energy of formation for metal oxides drops while that of carbon oxide increases (CO) or remains constant (CO₂) when temperature increases. This diagram is very useful to predict the temperature required for the carbothermal reduction of certain metal oxides or sulfides from the thermodynamic point of view. For any given metal oxide, it could be reduced by carbon when its free energy of formation is above that of either CO or CO₂ for certain temperature.

As a carbon based structure, crumpled GO nanosack may also be a good platform for producing metal nanostructures, which are not very stable during storage or are hard/sophisticated to produce with traditional synthetic routes. Zero valance iron (Fe⁰) nanoparticle is one of such examples. Due to its high reactivity, Fe⁰ NPs tend to oxidize during preparation and storage, making process control very difficult. In addition, they are highly magnetic and thus strongly aggregate in solution, which causes it very challenging to obtain good colloidal dispersion. Here we use G/Fe₃O₄ as a precursor to produce G/Fe⁰ as a demonstration and this method can in principle be applied to achieve similar goals with other iron based compounds. G/Fe₃O₄ nanosacks which are facilely produced can be used as a precursor to produce Fe⁰ NPs with carbon shells, which provide protection against oxidative passivation. Fe₃O₄ is expected to be reduced by the carbon sheets between 700 and 750 °C as the Ellingham diagram shows the free energy of formation of Fe₃O₄ intersects with those of CO and CO₂. Heating G/Fe₃O₄ at 700 °C in nitrogen flow for 2 h partially

reduced Fe_3O_4 into a mixture of FeO and Fe_3O_4 (Figure 5.6A and B) as the temperature is not enough high to fully convert Fe_3O_4 to Fe^0 ; however, heating at 800 °C for 2 h changes all Fe_3O_4 into Fe^0 (Figure 5.6C and D). The cargo nanosacks morphology remains the same as the $\text{G}/\text{Fe}_3\text{O}_4$ at the two temperatures. Higher temperature like 900 °C can produce similar Fe^0 phase structure with better crystallinity but the sacks carbon shell collapse and the Fe^0 particles fuse together to form large micron size particles.

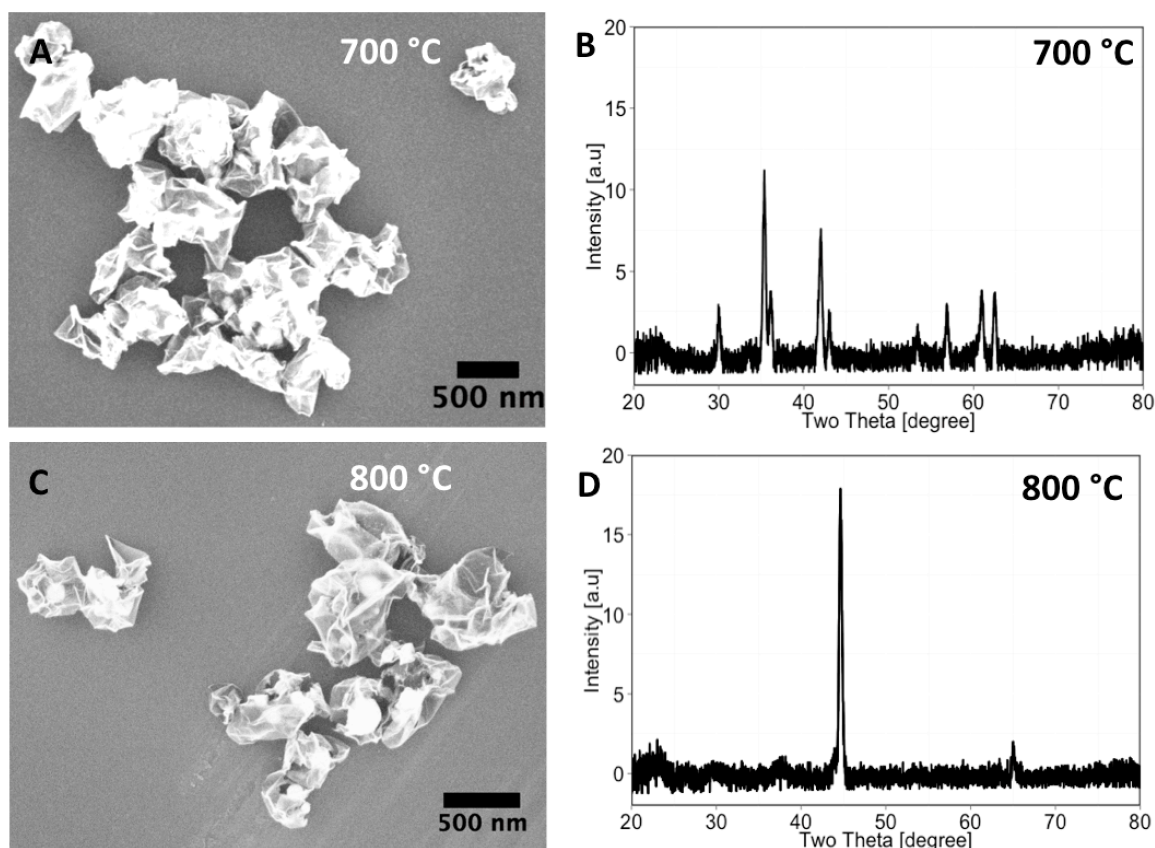


Figure 5.6 Carbothermal reduction by the graphene sheets for the synthesis of G/Fe^0 nanosacks using $\text{G}/\text{Fe}_3\text{O}_4$

Both 700 and 800 °C treatment of $\text{G}/\text{Fe}_3\text{O}_4$ for 2 hours maintains the nanosacks structure (A,C) 700 °C treatment for 2 h partially reduced the cargo Fe_3O_4 nanoparticles into a mixture of FeO and Fe_3O_4 (B), but 800 °C treatment for 2 h reduced all the cargo nanoparticles into Fe^0 nanoparticles. (D)

One interesting observation is that carbothermal reduction can even happen within the extremely short period of time when droplets fly through the furnace, which is less than 1 s.

The precursor FeOOH nanoparticles can be reduced to Fe₃O₄ if the aerosol process is done at 300 °C or above. Although 300 °C is enough to obtain magnetite, higher temperature is favorable for better crystallinity, but in the meantime the encapsulated particles become more porous as process temperature increases. Definitely, the volume of FeOOH particles will decrease when they transform to Fe₃O₄ due to the loss of inner water, and thus create porous structures. Previous studies reported that these pores could result in shrinkage of bare FeOOH NPs at 300 °C, and total structural collapse at 550 °C.²⁴ Whereas, the Fe₃O₄ particles wrapped inside rGO sacks do not shrink, instead those pores grow, connect and finally integrate into a single hollow, resulting in a hollow nanorod structure of Fe₃O₄ when using 500 °C for aerosol process. However, further increase to 600 °C will lead to structure damage and debris from broken particles can be observed, and then breakage or collapse happens with 700 °C aerosol process.

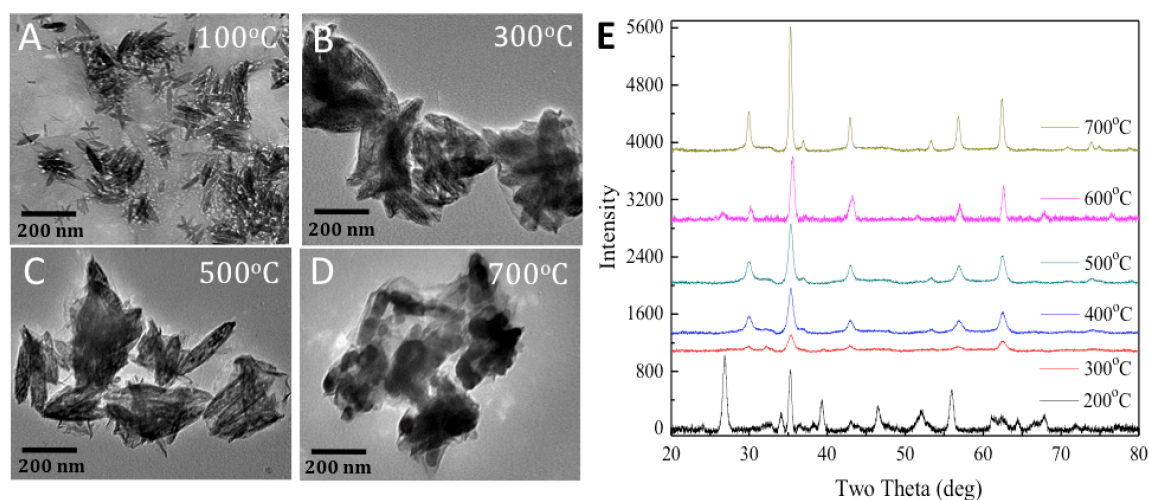


Figure 5.7 Changes of obtained iron-containing rGO nanosacks with increasing temperature for the thermal aerosol processing

(A-D) morphology changes in TEM images, (E) structure changes in XRD patterns. The XRD pattern at 200 °C is characteristic pattern of FeOOH and those of the rest are characteristic pattern of Fe₃O₄

5.4 Conclusion

These results show that graphene encapsulation in the aerosol phase is a useful technique for fabrication of multifunctional materials. Ensembles of simple unfunctional nanoparticles can be wrapped by rGO sheets in a non-chemically specific manner to produce multifunctional structures without the need for complex multistep chemical synthesis. A wide variety of nanoparticle cargoes can be encapsulated this way, including some that are colloidally unstable on their own but become stable through electrostatic association with GO sheets. In some cases, particle/sheet association becomes extensive and large heteroaggregates form that disrupts uniform aerosol formation and encapsulation. In these cases, high pH is shown to be the favorable operating regime where GO and most nanoparticle cargoes have common negative surface charge and thus disperse uniformly and independently. High pH also enhances the stability of some metal and metal oxide NPs to dissolution or oxidative dissolution.

This study also shows that graphene sacks in as-produced form are open to small solute exchange with their environment but can be partially sealed by addition of polymer filler to the nebulizing suspension. The partial sealing suggests potential for controlled release applications and their combination with multimodal imaging as demonstrated here toward the goal of GO nanosack-based theranostics. This study also demonstrates the multifunctional material concept by fabricating G/Au/Fe₃O₄ and G/BaTiO₃/Fe₃O₄ ternary particles as MRI/CT multimodal imaging probes. The G/Au/Fe₃O₄ probe shows both MRI and CT contrast in CMC gel suspensions at low concentrations ranging from 0.05 to 2000 µg/mL based on tests using clinical-scale scanners. While this first experiment demonstrated both MRI and CT contrast, further work is needed to develop multimodal

probes for specific applications. We envision the optimization of cargoes, by using smaller Fe_3O_4 nanoparticles for increased developed magnetization,^{46,47} and the use of smaller nanosacks with higher number densities for improved vascular penetration to tumors through the enhanced permeability and retention effect.

Finally, in addition to stabilizing the carbon shells of nanosacks, thermal treatment during or after the aerosol process is found to play a significant role in determining the composition or structure of the products. This could be due to the reductive property of carbon sheets at high temperature, the spontaneous phase/composition transformation of the encapsulated cargo material at different temperatures or both.

5.5 References

1. Cote, L. J.; Kim, F.; Huang, J., Langmuir-Blodgett assembly of graphite oxide single layers. *J. Am. Chem. Soc.* 2009, *131* (3), 1043-9.
2. Chen, Y. T.; Guo, F.; Jachak, A.; Kim, S. P.; Datta, D.; Liu, J. Y.; Kulaots, I.; Vaslet, C.; Jang, H. D.; Huang, J. X.; Kane, A.; Shenoy, V. B.; Hurt, R. H., Aerosol Synthesis of Cargo-Filled Graphene Nanosacks. *Nano Lett.* 2012, *12* (4), 1996-2002.
3. Ma, X. F.; Zachariah, M. R.; Zangmeister, C. D., Crumpled Nanopaper from Graphene Oxide. *Nano Lett.* 2012, *12* (1), 486-489.
4. Luo, J. Y.; Jang, H. D.; Sun, T.; Xiao, L.; He, Z.; Katsoulidis, A. P.; Kanatzidis, M. G.; Gibson, J. M.; Huang, J. X., Compression and Aggregation-Resistant Particles of Crumpled Soft Sheets. *ACS nano* 2011, *5* (11), 8943-8949.
5. Yue, J.; Jiang, X. C.; Yu, A. B., Experimental and theoretical study on the beta-FeOOH nanorods: growth and conversion. *J. Nanopart. Res.* 2011, *13* (9), 3961-3974.
6. Haynes, W. M., In *CRC Handbook of Chemistry and Physics*, Taylor and Francis: Boca Raton, FL, 2012; p 10 232.
7. Gomez-Navarro, C.; Weitz, R. T.; Bittner, A. M.; Scolari, M.; Mews, A.; Burghard, M.; Kern, K., Electronic transport properties of individual chemically reduced graphene oxide sheets. *Nano Lett.* 2007, *7* (11), 3499-3503.
8. Daou, T. J.; Pourroy, G.; Greneche, J. M.; Bertin, A.; Felder-Flesch, D.; Begin-Colin, S., Water soluble dendronized iron oxide nanoparticles. *Dalton T* 2009, (23), 4442-4449.
9. Kim, J.; Cote, L. J.; Kim, F.; Yuan, W.; Shull, K. R.; Huang, J. X., Graphene Oxide Sheets at Interfaces. *J. Am. Chem. Soc.* 2010, *132* (23), 8180-8186.

10. Liu, J. Y.; Hurt, R. H., Ion Release Kinetics and Particle Persistence in Aqueous Nano-Silver Colloids. *Environ. Sci. Technol.* 2010, 44 (6), 2169-2175.
11. Miao, A. J.; Zhang, X. Y.; Luo, Z. P.; Chen, C. S.; Chin, W. C.; Santschi, P. H.; Quigg, A., Zinc Oxide Engineered Nanoparticles Dissolution and Toxicity to Marine Phytoplankton. *Environ. Toxicol. Chem.* 2010, 29 (12), 2814-2822.
12. Midander, K.; Cronholm, P.; Karlsson, H. L.; Elihn, K.; Moller, L.; Leygraf, C.; Wallinder, I. O., Surface Characteristics, Copper Release, and Toxicity of Nano- and Micrometer-Sized Copper and Copper(II) Oxide Particles: A Cross-Disciplinary Study. *Small* 2009, 5 (3), 389-399.
13. Lastoskie, C.; Gubbins, K. E.; Quirke, N., Pore-Size Heterogeneity and the Carbon Slit Pore - a Density-Functional Theory Model. *Langmuir* 1993, 9 (10), 2693-2702.
14. Samson, E.; Marchand, J.; Snyder, K. A., Calculation of ionic diffusion coefficients on the basis of migration test results. *Mater. Struct.* 2003, 36 (257), 156-165.
15. Kircher, M. F.; Willmann, J. K., Molecular Body Imaging: MR Imaging, CT, and US. Part I. Principles. *Radiology* 2012, 263 (3), 633-643.
16. Sana, B.; Johnson, E.; Sheah, K.; Poh, C. L.; Lim, S., Iron-based ferritin nanocore as a contrast agent. *Biointerphases* 2010, 5 (3), Fa48-Fa52.
17. Sana, B.; Poh, C. L.; Lim, S., A manganese-ferritin nanocomposite as an ultrasensitive T-2 contrast agent. *Chem. Commun.* 2012, 48 (6), 862-864.
18. Yan, G.-P.; Robinson, L.; Hogg, P., Magnetic resonance imaging contrast agents: Overview and perspectives. *Radiography* 2007, 13, Supplement 1, e5-e19.
19. Tasis, D.; Tagmatarchis, N.; Bianco, A.; Prato, M., Chemistry of carbon nanotubes. *Chem. Rev.* 2006, 106 (3), 1105-1136.
20. Lacerda, L.; Bianco, A.; Prato, M.; Kostarelos, K., Carbon nanotubes as nanomedicines: From toxicology to pharmacology. *Adv. Drug Del. Rev.* 2006, 58 (14), 1460-1470.

21. Robinson, J. T.; Tabakman, S. M.; Liang, Y. Y.; Wang, H. L.; Casalongue, H. S.; Vinh, D.; Dai, H. J., Ultrasmall Reduced Graphene Oxide with High Near-Infrared Absorbance for Photothermal Therapy. *J. Am. Chem. Soc.* 2011, *133* (17), 6825-6831.
22. Kam, N. W. S.; O'Connell, M.; Wisdom, J. A.; Dai, H. J., Carbon nanotubes as multifunctional biological transporters and near-infrared agents for selective cancer cell destruction. *Proc. Natl. Acad. Sci. U. S. A.* 2005, *102* (33), 11600-11605.
23. Wu, B. B.; Liu, Y. A.; Rose-Petruck, C.; Diebold, G. J., X-ray spatial frequency heterodyne imaging. *Appl. Phys. Lett.* 2012, *100* (6).
24. Piao, Y.; Kim, J.; Na, H. B.; Kim, D.; Baek, J. S.; Ko, M. K.; Lee, J. H.; Shokouhimehr, M.; Hyeon, T., Wrap-bake-peel process for nanostructural transformation from beta-FeOOH nanorods to biocompatible iron oxide nanocapsules. *Nat Mater* 2008, *7* (3), 242-247.

CHAPTER 6 SUMMARY OF THESIS

GO-based materials as the most promising graphene materials for large-scale application deserve more study on their properties. Better understanding on their chemical and thermochemical properties could be important for clarifying controversies on their health and safety issues, which are essential for these materials' successful uptake by the industry.

The energetic nature of GO is ultimately due to the exothermic nature of its reduction, which is determined by its special chemical structure, more specifically, the large amount of epoxy groups. Heating condition, storage form and presence of impurities can change its chemical reactivity or the heat and mass transfer efficiency during the process. Explosion is the result of a self-propagating reaction confined in a small region of GO by limited heat and mass transfer. Redox activity of GO-based materials largely determines their ability to cause oxidative stress in biological system, which is an important mechanism to toxicity. GO-based materials can be either pro- or anti- oxidative depending on their biological targets. They can inhibit ROS generation as well as scavenge existing ROS. Due to its unique chemical properties, GO can also be used to build 3D self assemblies which can simultaneously encapsulate multiple types of cargo, serve as protective antioxidant layer to remediate oxidative damage, synthesize and stabilize the highly reactive Fe^0 NPs without hazardous reductants and other types of applications.

This study provides insights for developing safer and more environmentally responsible processing techniques for a couple of GO-based materials with different chemical properties. It would be a good source for manufacturers to refer to in their processing practices of GO-based materials.