NANOPOROUS LAYERED GRAPHENE HYDROGEL FOR CONTROLLED DRUG DELIVERY

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Declaration

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Abstract

Graphene-related materials with tuneable pore sizes in the nanoscale range offer the potential to address significant challenges in biomolecule separation, controlled delivery of drugs, selective biosensor, rechargeable batteries, supercapacitors and solar cells. Layered assemblies of graphene-related sheets with physical and chemical cross-linkers between the sheets have been recognized as one possible strategy for making such nanoporous materials. However, current approaches give very limited control over the pore size distribution, particularly with regards control of the mean pore size and the degree of spread around it.

This work particularly outlined the design, synthesis and characterization of a nanoporous layered graphene hydrogel produced via peptide-mediated self-assembly of reduced graphene oxide (rGO). The peptides have been designed using molecular dynamics (MD) simulation to self-assemble the rGO sheets with a desired inter-sheet spacing (pore size). The hydrogel material was synthesized and characterized using a range of methods to demonstrate the desired pore size is achieved.

In the second body of this work, the rGO binding peptide hydrogel, denoted rGOPH, showed to be a promising candidate for the controlled delivery of an anti-cancer drug. In particular, it was shown that the rGOPH has a high doxorubicin (DOX) loading capacity achieved through physical adsorption within its nanoporous structure. Design of experiments (DoE) and statistical analysis on different preparation parameters revealed that pore size and drug loading capacity are tuneable.

In the final part of the work, a desirable pH-dependant drug release properties was shown by rGOPH nominating such hydrogels as promising candidates for cancer therapy. In addition, the hydrogel materials exhibited a high biocompatibility to the healthy cells for their attachments and proliferation. The cytotoxicity of the hydrogel materials demonstrated to be low.

The work reported in this thesis has provided new computational and experimental understanding for fabrication of graphene based nano-constructs with tuneable pore size as

well as new methodologies and approaches. Although the focus was only on one designed peptide, the design and methodologies developed here are quite potent and, therefore, lay the foundations for fabrication of nanoporous graphene based materials of virtually any pore size to suit the needs of users in broader applications (such as nanomedicines, nanobiotechnology, nanoelectronics, biosensors and biomolecular and nanoparticle separations).

Achievements

Two patents were achieved from this work:

1) Compositions comprising self-assembled carbon based structures and related methods. A. P. Patent. **AU2014/900273**.

2) Self-assembled carbon based structures and related methods. **PCT/AU2015/000034**.

This work was presented in conferences with the following titles:

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3) "Molecular Modelling of Protein Adsorption: From Fundamentals to Design." FOA11 (the 11th International Conference on the Fundamentals of Adsorption), (2013) Baltimore, Maryland, USA.
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Abbreviations

ANOVA	Analysis of variance
AFM	Atomic force microscopy
BJH	Barrett-Joyner-Halenda
CPT	Camptothecin
CB	carbon black
CNT	Carbon nanotube
CCD	Central composite design
CCFD	Central composite face centered design
CTAB	Cetyltrimethylammonium bromide
CVD	Chemical vapor deposition
CCG	Chemically converted graphene
CF	Ciprofloxacin
CV	Coefficient of variation
DOE	Design of experiments
DMSO	Dimethyl sulfoxide
DTAB	Dodecyltrimethylammonium bromide

ds-DNA	Double stranded
DOX	Doxorubicin
DDS	Drug delivery systems
EthD-1	Ethidium homodimer-1
FD4,10 and20	Fluorescein isothiocyanate–dextran (4, 10 and 20 KD)
FTIR	Fourier transform infrared
Glu	Glutamic acid
Gly	Glycine
AuNP	Gold nanoparticle
g-C3N4	Graphene based carbon nitride
GO	Graphene oxide or graphite oxide
GS	Graphene sheet
HOPG	Highly ordered pyrolytic graphite
hFOB	Human fetal osteoblast
HOG	Human oligodendroglia
LBL	Layer by layer

LCST	Lower critical solution temperature
M-LBL	Manual layer-by-layer
MSCs	Mesenchymal stem cells
MD	Molecular dynamics
MM	Molecular mechanics
MC	Monte Carlo
DMF	N,N-dimethylformamide
NG	Nitrogen doped graphene
NMP	N-methyl-2-pyrrolidone
Phe	Phenylalanine
PMAA	Poly (methacrylic acid)
P(AA-co-AM)	Poly(acrylic acid-co-acrylamide)
PMVE	Poly(methylvinylether)
DEAM	Poly(N,N'-diethylacrylamide)
PAcrNPP	Poly(N-acryloyl-N'-Propylpiperazine)
PNIPAAm	Poly(N-isopropylacrylamide)

PVA	Poly(vinyl alcohol)
PAA	Polyallylamine
PANI	Polyaniline
PDMS	Polydimethylsiloxane
PEG	Polyethylene glycol
PEI	Polyethyleneimine
PET	Polyethyleneterephthalate
PPy	polypyrrole
PSD	Pore size distribution
PPD	p-phenylenediamine
QM	Quantum mechanics
QSDFT	Quenched Solid Density Functional Theory
ROS	Reactive oxygen species
rGO	Reduced graphene oxide
rGOH	Reduced graphene oxide hydrogel
RSM	Response surface method

rGOPH	rGO binding peptide hydrogel
SEM	scanning electron microscopy
SA	Self-assembly
SiC	Silicon carbid
ss-DNA	Single stranded DNA
SWCNTs	Single wall carbon nanotubes
SD	Standard deviation
SA	Succinic acid
TPA	Terephthalic acid
THF	Tetrahydrofuran
TEM	Transmission electron microscopy
3D	Tri-dimensional
Trp	Tryptophan
2D	Two-dimensional
Tyr	Tyrosine
UHV	Ultra high vacuum

UV-vis	Ultraviolet-visible
UCST	Upper critical solution temperature
VACNTs	vertically aligned CNTs
VMD	Visual molecular dynamics
VPTT	Volume phase transition temperature
WAXRD	Wide Angle X-ray Diffraction
XPS	X-ray Photoelectron Spectroscopy

Nomenclatures

M_∞	Final amount of molecule released after an infinite time
M_t	Cumulative amount of drug released at time *t*
D	Drug diffusion coefficient
L	Thickness of the drug-releasing implant
pKa	Acid dissociation constant,
$\Delta\omega$	Raman shift (in cm^{-1}), is the, and
λ_0	Excitation wavelength
λ_1	Raman spectrum wavelength
P/P0	Relative pressure (-)
d002	Interlayer spacing of (002) face (nm)
λ	Wavelength (nm)
E_b	Electron binding energy
d_{hkl}	Interplanar spacing of planes (between the layers of atoms)
θ	Bragg angle between the incident x-ray beam and the surface of crystal
A	Measured absorbance

I_{in}	Intensity of the incident radiation
I_{out}	Transmitted intensity
L	Path length of light travels through the cuvette
ε	Molar extinction coefficient
c	Sample concentration
$W_{initial\ DOX}$	Weight of DOX initially added
$W_{final\ DOX}$	Weight of DOX left in the cuvette after 24 hrs
$W_{hydrogel}$	Weight of rGOH and rGOPH samples
Y_i	Predicted response (dependent variable
X_i	Independent variables
X_iX_j	Variables interactions
β_0	Constant coefficient
β_i	Coefficients for the linear effects
β_{ii}	Coefficients for the quadratic effects
β_{ij}	Coefficients for the interaction effects
ε	Standard error

k	Number of independent variables
p(F)	Probability of Fisher's F-test
k_1	Lagergren rate constant of adsorption (min^{-1})
q_e	Ultimate adsorption capacity
q_t	Adsorption capacity at time *t*
k_2	Pseudo second-order rate constant of adsorption (g mg^{-1} min)
k_{ipd}	Rate constants of intra-particle diffusion (mg g^{-1} $min^{-0.5}$)

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