Diamond micro-cantilevers as transducers for olfactory receptors - based biosensors

Raafa Manai, Dounia Kamouni-Belghiti, Marie-Annick Persuy, M. Habchi, Lionel Rousseau, M. Possas Abreu, P. Bergonzo, Edith Pajot, Guenhaël Sanz, E. Scorsone

ICONAN 2016

International Conference on NANOMEDICINE And NANOBIOTECHNOLOGY

BOOK OF ABSTRACTS

University Pierre and Marie Curie, Paris

Sept 28-30, 2016

PremC
Conferences, Events & Workshops

premc.org/iconan2016

particle works

materials at peak performance

- Batch to batch consistency
- Precision engineered nanoparticles
- Supplied with full characterization data

Visit Particle Works on Patio 25!

www.particle-works.com

nanolane

EYES ENTERING THE NANOWORLD

N-Lab Station

New label-free surface analysis equipment

- Live nanoscale imaging
- Direct quantitative analysis
- Multiplex surface interaction study
- Thermal and fluidic control
- Mode: Air/Liquid/Gas
- Microfluidic compatibility

Committee

Chairwoman

Ane-Marie Caminade, University of Toulouse

Scientific Committee

Albana Halili, Epoka University
William Waissmann, Oswaldo Cruz Foundation / São Paulo State University
Christophe Ramseyer, Université de Bourgogne Franche-Comté
Iwona Cicha, University Hospital Erlangen, Section of Experimental Oncology and Nanomedicine (SEON)
Niko Hildebrandt, Université Paris-Sud
Jeffrey Keelan, University of Western Australia
Mariano Venanzi, University of Rome 'Tor Vergata'
Maria Blanco-Prieto, University of Navarra
Bjørn Torger Stokke, The Norwegian University of Science and Technology, NTNU
Fabio Pastorino, IRCCS Istituto G. Gaslini
Suna Timur, Ege University
Wafa Al-Jamal, UEA
Serge Ravaine, Bordeaux University
Andre G Skirtach, Ghent University
Vadim Kessler, SLU
Ana Belén Gonzalez-Guerrero, Catalan Institute on Nanoscience and Nanotechnology
Kenneth E. Gonsalves, IIT Mandi Himachal Pradesh 175001
Luis Liz-Marzán, CIC biomaGUNE
David Fairen-Jimenez, University of Cambridge
Rongjun Chen, Imperial College London
Florence Gazeau, Paris Diderot University
Sijumon Kunjachan, Harvard Medical School
Claire Billotey, Université de Lyon, UJM, EMR 3738
Stéphane Roux, Institut UTINAM, Université de Franche-Comté
Nadine Millot, Université de Bourgogne Franche-Comté
Christophe Moreau, Institut de Biologie Structurale, Grenoble
Casuso Ignacio, INSERM U1006 - BioAFM Lab
James N Sturgis, Aix-Marseille Université
Eric Finot, University of Burgundy
Eric Lesniewska, University of Bourgogne / ICB UMR CNRS 6303
Edith Pajot-Augy, INRA
Abdelhamid Errachid, The Institute of Analytical Sciences
Ibtisam E Tothill, Cranfield University
Gaëlle Offranc Piret, U1205 INSERM-UGA-CHU
Rogério Gaspar, University of Lisbon
Thierry Pécot, INRIA Rennes - Bretagne Atlantique
Emilie Roger, Angers University
Nicolas Bertrand, Université Laval
Fabien Picaud, Université de Bourgogne Franche-Comté
Sebastien Balme, Université de Montpellier
Etienne Dague, CNRS
Min-Hui Li, Chimie ParisTech - ENSCP
Ciro Chiappini, Imperial College London
Fernando Torres Andón, Istituto Clinico Humanitas
Ana Luísa Cardoso, FCA – State University of Campinas
Nejat Düzgüneş, University of the Pacific School of Dentistry
Dongyang Li, University of Alberta

Seda Kizilel, Koc University
Zofia Lipkowska, Institute of Organic Chemistry, Polish Academy of Sciences

Plenary Speakers

Anne-Marie Caminade, University of Toulouse
Ick Chan Kwon, Korea Institute of Science and Technology
Dennis E. Discher, University of Pennsylvania
Jackie Ying, Institute of Bioengineering and Nanotechnology,
Rabah Boukherroub, Lille 1 University
Fabian Kießling, ExMI – Experimental Molecular Imaging
Claire Wilhelm, Paris Diderot University
Kenneth A. Dawson, University College Dublin
Christian Serre, ENS-ESPCI-PSL-CNRS
Philippe Barthélémy, Bordeaux Segalen University
Elias Fattal, University of Paris-Sud
Kostas Kostarelos, University of Manchester

Organizing Committee

PremC

Table of Contents

Theragnostic Nanoparticles for Drug Delivery Systems

Wednesday, 28th September - 09:05 - Plenary Speeches - Amphitheatre 25 - Oral presentation - Abstract ID: 564

Prof. Ick Chan Kwon [1]

1. *Korea Institute of Science and Technology*

Recently, nanoparticles have received a great interest an application for diagnosis and therapy. Since nanoparticles possess intrinsic features that are often required for drug delivery system and diagnosis, they have potential as platforms for integrating imaging and therapeutic functions, simultaneously. Especially, molecular imaging with theragnostic nanoparticles makes it possible not only to provide useful information for monitoring drug delivery, drug release, and therapeutic efficacy of drug, but also to determine whether the patients are likely to respond to a therapy. To achieve these goals, a variety of imaging techniques have been used, including near-infrared fluorescent (NIRF) imaging, magnetic resonance imaging (MRI) and nuclear imaging (SPECT and PET). Imaging and monitoring of nanoparticles after systemically administered in living systems play key roles in the development of theragnostic nanoparticles to optimize their physicochemical properties. It has become clear that imaging drug delivery can assist in analyzing the drug delivery and in predicting the therapeutic efficacy of cancer-targeted nanoparticles. This presentation will highlight our recent advances that have been made in the development of multifunctional nanoparticles and the applications of these nanoparticles into theragnostic nanomedicine.

Magnetic nanoparticles in cell therapies: from combined approaches in cancer treatment to magnetic tissue engineering.

Wednesday, 28th September - 09:40 - Plenary Speeches - Amphitheatre 25 - Oral presentation - Abstract ID: 551

Prof. Claire Wilhelm [1]

1. *CNRS and Paris Diderot University, laboratory Matière et Systèmes Complexes.*

To overcome some of the limitations of current cell therapies tools, new strategies have emerged since the advent of nanotechnology in medicine.
In cancer therapy, thermal treatments (magnetic hyperthermia or photothermal therapy mediated by magnetic or plasmonic nanoparticles) have provided noninvasive means of heating cells at therapeutic levels. However, while the ultimate target for nanoparticle-mediated photothermal therapy is the cancer cell, heating performance has not previously been evaluated inside the cells. In the attempt to bridge this gap, we provided the first thermal measurements mediated by magnetic [1] or plasmonic [2] nanoparticles inside cancer cells, in vitro or in vivo in the tumor environment. The ultimate goal of nanotherapies is anyway to improve the efficacy and combat the tumour from within. We proposed combined nanotherapeutic concepts [3-6] based on magnetothermal, photothermal, and photodynamic therapies which led to complete cancer cell destruction in vitro and complete tumor ablation in vivo.
While magnetic nanoparticles are increasingly used as clinical agents for imaging and therapy, their use as a tool for tissue engineering opens up challenging perspectives that have rarely been explored. Our strategy has been to take advantage of magnetic nanoparticles internalization to create thick, organized, purely cellular 3D tissue structures [7,8] , that can be stimulated on demand [9].
The use of nanoparticles for cancer cell therapies or tissue engineering raise more general issues of nanoparticles biosafety, once internalized in cells. Yet the nanoparticles long-term tissular fate is poorly documented. We have developed original magnetic techniques to follow the fate of iron oxide nanoparticles and their assimilation within a living tissue, and evidenced that a massive biodegradation can occur at the endosome site.
[1] Biomaterials 24, 6400–6411 (2014)
[2] Advanced HealthCare Materials, 10.1002/adhm.201501035 (2016)
[3] ACS Nano, 10, 2436-46 (2016)
[4] Nanoscale, 7, 18872-18877 (2015)
[5] Nanomedicine. 10, 2797 (2015)
[6] ACS Nano, 9, 2904-2916 (2015)
[7] Advanced Materials. 25, 2611-2616 (2013)
[8] Integrative Biology, 7, 170-177 (2015)
[9] Phys Rev Lett, 114, 098105 (2015).

Nano-antibiotics: A rational design of functional nanoparticles to combat bacterial infection

Wednesday, 28th September - 10:45 - Plenary Speeches - Amphitheatre 25 - Oral presentation - Abstract ID: 542

Dr. Rabah Boukherroub [1]

1. *CNRS, Lille University*

Complications related to infectious diseases have significantly reduced, particularly in the developed countries, due to the availability and use of a wide variety of antibiotics and antimicrobial agents. However, excessive use of antibiotics and antimicrobial agents increased the number of drug resistant pathogens, and this has resulted in a significant threat to public health. The inexorable rise in the incidence of antibiotic resistance in bacterial pathogens, coupled with the low rate of emergence of new clinically useful antibiotics, has refocused attention on finding alternatives to overcome antimicrobial resistance. Novel strategies aiming to reduce the amount of antibiotics, but able to prevent and treat animal and human infections should be investigated, evidenced and approved.

Among the various approaches, the use of nanotechnology (engineered nanoparticles) is currently the most promising strategy to overcome microbial drug resistance. Due to their small size, nanoparticles can surmount existing drug resistance mechanisms, including decreased uptake and increased efflux of drug from the microbial cell, biofilm formation, and intracellular bacteria. Moreover, loading multiple antimicrobial agents on the same nanoparticle makes the development of resistance unlikely. Finally, nanoparticles can target antimicrobial agents to the site of infection, so that higher doses of drug are given at the infected site, thereby overcoming resistance.

Despite considerable recent progress in the understanding of the mechanisms underlying bacterial infections, and in the development of nanostructured materials displaying antibacterial properties and activity against biofilms, the quest to design and fabricate new antibacterial nanostructures remains a high research priority.

In this presentation, after an overview on the different nanomaterials possessing antimicrobial activities, I will discuss the use of nanosized drug carriers to efficiently administer antibiotics by improving their pharmacokinetics and bioaccumulation, while reducing the adverse effects of antibiotics.

Considerations on the design of nanoparticles for diagnostic, theranostic and therapeutic purposes

Wednesday, 28th September - 11:25 - Plenary Speeches - Amphitheatre 25 - Oral presentation - Abstract ID: 524

Prof. Fabian Kiessling [1]

1. *RWTH Aachen University*

Nanoparticles are frequently suggested for medical use. However, often basic considerations on its pharmacokinetic properties are not taken into account. Additionally, its optimal characteristics have to be very different if they are considered for diagnostic or therapeutic purposes.

Nanoparticles >5 nm tend to be removed by the RES. Thus, tissues belonging to the RES like liver spleen and lymph nodes can be targeted with such nanoparticles. Furthermore, if phagocyting cells migrate to pathological sites, the nanoparticles will also be accumulated, which has been shown e.g. for (U)SPIO. Phagocyting cells can also be labelled ex vivo with diagnostic nanoparticles, which opens great perspectives for cell tracking and the imaging of tissue engineered transplants.

Adding stealth properties to the nanoparticles increases their circulation time giving them more time to extravasate in tissue with high vessel permeability. This so called EPR based accumulation is the basis for most tumor targeted nanomedicines. However, the therapeutic benefit over small probes is often only moderate since EPR is variable among patients and even heterogeneous within the same tumor. Here, theranostic agents and companion diagnostics can help to preselect patients and to individualize therapy. In addition, in tumors larger nanoparticles tend to accumulate just outside the vasculature, do hardly penetrate the stroma and thus do not reach the cancer cells. Thus, a refined balance between accumulation and penetration may be therapeutically superior over just maximal accumulation. In this context, active targeting does only marginally help since it does not improve nanoparticle distribution and accumulation but just retention. Active targeting becomes more evident for small nanoparticles (< 5 nm) showing good penetration and rapid exchange between the tissue compartments but insufficient retention. Additionally, these are the ones that are most suited for molecular imaging purposes as well.

Thus, the intended medical application should route the decisions about design of nanoparticles and all aspects relevant to its in vivo application including the expected superiority over existing clinical gold standards should be considered from beginning on. Following this conduct, many failures in the transition from in vitro to in vivo application can be avoided.

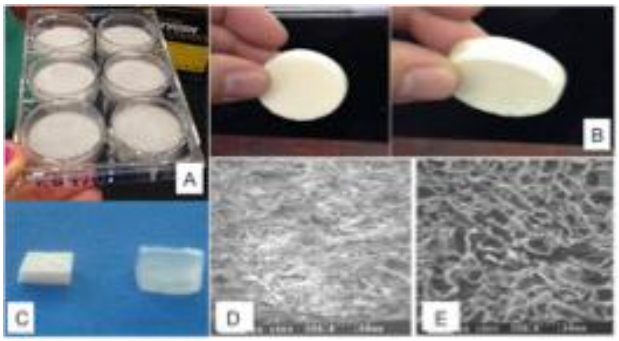
Chitosan-Collagen hybrid 3D-scaffolds as potential biomaterials for tissue engineering

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 81

Ms. Hilary Urena [1]***, Dr. Marianelly Esquivel-Alfaro*** [2]***, Dr. Sergio Madrigal-Carballo*** [1]

1. *National Center for Biotechnological Innovations (CENIBiot), CeNAT-CONARE,* ***2.*** *Polymers Research Laboratory (POLIUNA), National University*

Introduction. Chitosan has been applied to promote extracellular matrix (ECM) formation in tissue regenerative therapy. The superior tissue compatibility of chitosan may primarily be attributed to its structural similarity to glycosaminoglycan in ECM. Chitosan has been reported to be biocompatible, bio-absorbable and particularly, is considered a good wound-healing accelerator. Dermis and scaffolds made from chitosan exhibit weak antigenicity, biodegradability, and superior biocompatibility (hemostatic and cell-binding properties) by comparison to the synthetic polymers, such as poly(lactic acid) (PLA), poly(glycolic acid) (PGA), and polyethylene terephthalate (PET). As a scaffold, chitosan-based materials in the form of a sponge have been considered the most popular 3D-scaffolds for dermal regeneration. Of the many scaffold materials being investigated, collagen has been shown to have many advantageous features. Highly porous collagen lattice sponges have been used to support in vitro growth of many types of tissues. Methods. We isolated chitosan from native shrimp waste streams and collagen from tilapia aquaculture waste by-products. Hybrid 3D-scaffold biomaterials were successfully obtained by mixing chitosan with collagen at different molar ratios. Chitosan-collagen hybrid composites were formulated as 3D sponge-like scaffolds, applying previously developed methodologies involving solvent casting and freeze-drying. Results. Chitosan-collagen hybrid 3D-scaffolds were characterized according to its water uptake capacity, thermal behavior (DSC) and morphology (SEM). Discussion. Chitosan-collagen hybrid 3D-scaffolds showed improved stability, higher active compound loading capacity, better release properties, improved cell uptake, greater porosity, tensile strength and increased thermal stability as compared to current synthetic scaffolds. These hybrid nanostructured biomaterials will be suitable for the incorporation of active ingredients that may potentiate its application as dressings, cell culture scaffolds We are currently starting cell growth studies on the scaffolds using model epithelial cells.



Cht-cgn scaffolds.jpg

Corona interactome: a key for deciphering protein adsorption kinetics on silica nanocarriers

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 554

Mr. Cédric Pisani [1], ***Mr. Jean-charles Gaillard*** [2], ***Mr. Michaël Odorico*** [3], ***Dr. Jeff Nyalosaso*** [1], ***Dr. Clarence Charnay*** [1], ***Dr. Yannick Guari*** [1], ***Prof. Joël Chopineau*** [1], ***Prof. Jean-marie Devoisselle*** [1], ***Dr. Jean Armengaud*** [2], ***Dr. Odette Prat*** [4]

1. *Université de Montpellier,* ***2.*** *CEA, IBITECS,* ***3.*** *CEA, ICSM,* ***4.*** *CEA, BIAM*

Magnetic mesoporous silica nanoparticles (M-MSNs) represent promising targeting tools for cancer diagnostic and therapy. In biological systems, nanoparticles interact with proteins and form a layer named "corona" that drives their biological fate and toxicity. The corona around NP creates a new nano-object, whose interactions with living cells are different from those induced by the pristine NPs. We have investigated the behavior of adsorbed proteins around M-MSNs with two biological fluids, fetal bovine and human sera. The first one is interesting for in vitro toxicology and the second for in vivo diagnosis and therapy. Thereby, after a quantification of protein adsorption on these M-MSNs by BCA assay over time, we qualified these adsorbed proteins by high throughput comparative proteomics during long term kinetics. We observed an increase of the protein layer with both serum types from 30 seconds to 7 days of contact by both techniques. During this kinetics, the growth of the protein corona began instantly after NP immersion. We thus detected 90 to 128 and 134 to 153 distinct adsorbed proteins for the bovine and human corona, respectively. By using computational biology tools and protein-protein interaction databases, we have highlighted three major clusters of protein behavior in contact with nanoparticles. We demonstrated precisely how the protein network builds up within the corona during the experimental kinetics resulting in a multiparametric representation of the “corona interactome”. This original approach may allow understanding how NPs interact with biological fluids and provide some clues for the design of stealth nanocarriers.

Design of mesoporouse Fe3O4/MS nanoparticles as drug delivery platform of prednisolone

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 88

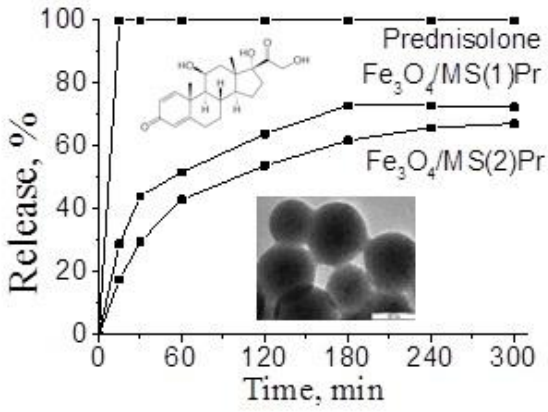
Mrs. Ivalina Trendafilova [1], ***Prof. Margarita Popova*** [2], ***Prof. Agnes Szegedi*** [3], ***Prof. Judith Mihály*** [3], ***Prof. Denitsa Momekova*** [4], ***Prof. Georgi Momekov*** [4], ***Dr. Irina Nikolova*** [4], ***Mr. Lyubomir Marinov*** [4]

1. *Institute of Organic Chemistry with Centre of Phytochemistry, Bulgarian Academy of Sciences, Sofia,* ***2.*** *Institute of Organic Chemistry with Centre of Phytochemistry, Bulgarian Academy of Sciences, 1113 Sofia,* ***3.*** *Research Centre for Natural Sciences, Institute of Materials and Environmental Chemistry, Hungarian Academy of Sciences, 1117 Budapest,* ***4.*** *Faculty of Pharmacy, Medical University of Sofia*

Interest towards mesoporous silicates as drug carries goes back to 2001, when application of MCM-41 type mesoporous silica for controlled delivery of drugs was published for the first time. Mesoporous silicates (MS) are characterized by narrow distribution of pores with a controlled size, large pore volume, high specific surface area (□700 m2/g), and good chemical and thermal stability. A new push towards the development of antitumor drug-delivery systems is given by the Fe3O4/MS mesoporous composites with different structures. By integration of mesoporous silica nanostructures with magnetic nanocrystals, original drug-delivery systems can be obtained permitting selective supply of the drug to the targeted organ or tissue of the body. Glucocorticoids i.e. prednisolone have been used in clinical oncology for more than three decades because their anti-inflammatory action additionally influences the oncological therapy.

In the present study we developed prednisolone loaded Fe3O4/MS nanoparticles. In-vitro release properties of the obtained delivery systems were studied in respect to their possible application in anti-cancer therapy. Fe3O4/MS nanoparticles with spherical morphology, small particle sizes (80 nm) and high surface area (□800 m2/g) were synthesized and loaded with prednisolone by incipient wetness impregnation. The Fe3O4/MS nanoparticles and drug formulations were characterized by XRD, N2 physisorption, TG analysis and ATR-FT-IR spectroscopy. In-vitro drug release study was performed into phosphate buffer (pH = 7) at 37oC. Loading of prednisolone on Fe3O4/MS nanoparticles resulted in high loading capacities. In-vitro release process at showed controlled prednisolone release. Spectroscopic data suggest the formation of a weak bond between silanols of the mesoporous Fe3O4/MS nanoparticles and prednisolone molecules. The cytotoxic potential of the presented mesoporous nanoparticles was tested in a panel of human malignant cell lines in-vitro. The results clearly indicate that the nanoparticles are practically devoid of toxicity in the tested concentration range. The in-vivo anti-inflammatory activity of prednisolone loaded mesoporous Fe3O4/MS nanoparticles vs. free drug was compared in murine collagen-induces arthritis. The results show that prednisolone encapsulation into mesoporous nanoparticles did not compromise its intrinsic pharmacological activity.

Acknowledgements Financial support from the Bulgarian-Hungarian Inter-Academic Exchange Agreement and the program for career development of young scientists, BAS, ДФНП 191/14.05.2016 is greatly acknowledged.


Release, %
Time, min
Prednisolone
Fe3O4/MS(1)Pr
Fe3O4/MS(2)Pr
0
20
40
60
80
100
0
60
120
180
240
300

Abstr.jpg

Cytotoxic Evaluation of Irinotecan-loaded PLGA-PEG-retinoic acid Nanomicelles on HT-29 Cancer Cell Line

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 218

Dr. Parnian Maghzi [1], ***Dr. Jaber Emami*** [1], ***Dr. Farshid Hasanzadeh*** [1], ***Dr. Hojjat Sadeghi*** [1]

1. *isfahan university of medical science*

In the present study PLGA-PEG and PLGA-PEG-retinoic acid (RA) which had already been synthesized in our laboratory were used to prepare targeted (PLGA-PEG-RA) and non-targeted (PLGA-PEG) polymeric micelles by thin-film hydration method to target irinotecan to colorectal cancer cell (HT-29). Cytotoxic effect of irinotecan solution, non-targeted irinotecan-loaded nanomicelles, targeted irinotecan-loaded nanomicelles, and blank nanomicelles were evaluated on HT-29 cell line using MTT assay. Untreated cells were taken as the negative control and the blank culture medium was used as the control. Cells were grown in RPMI 1640 medium and incubated with samples for 72 h at the equivalent irinotecan concentrations of 1 to 8 µg/ml. Cell viability for each sample was then calculated. Blank micelles did not show any measurable toxicity on HT-29 cell line indicating the safety of PLGA-PEG-RA as a potential drug carrier for irinotecan to the tumor cells. Toxicity of irinotecan and irinotecan-loaded nanomicelles were significantly increased in a drug concentration dependent manner. Non-targeted irinotecan-loaded nanomicelles showed greater cytotoxicity than free irinotecan while the cell toxicity of targeted irinotecan-loaded nanomicelles was significantly greater than both free irinotecan and non-targeted irinotecan-loaded nanomicelles. IC50 values of drug-loaded PLGA-PEG and drug-loaded PLGA-PEG-RA nanomicelles were significantly lower than those of the free drug. Irinotecan-loaded targeted nanomicelles exhibited lowest IC50 values as compared to free irinotecan and non-targeted nanomicelles. More cytotoxicity of non-targeted nanomicelles compared to free drug might be due to the presence of PEG on the surface of nanomicelles forming a brush-like shell that stretches away from the core reducing micelle aggregations and increasing cellular uptake. Drug-loaded PLGA-PEG-RA nanomicelles indicated highest toxicity compared to non-targeted nanomicelles which could be attributed to the receptor-mediated endocytosis and the nucleus-directed drug targeting of the nanoparticles. Our results indicate that the RA-conjugated nanomicelles could have great potential for targeted therapy of irinotecan, may reduce irinotecan therapeutic dose and consequently reduces adverse effects of this drug.

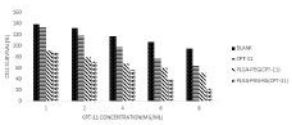


Fig. ht-29 cell survival.jpg

PREPARATION, CHARACTERIZATION AND RADIOLABELED OF VENLAFAXINE-PLGA NANOPARTICLES WITH 99mTC FOR BRAIN DELIVERY

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 115

Ms. María Dolores Cayero [1]***, Ms. ROCÍO IGLESIAS JEREZ*** [2]***, Mr. Roque Salazar Cabrera*** [3]***, Ms. Isabel Borrego Dorado*** [2]***, Dr. Lucía Martín-Banderas*** [1]

1. *Pharmacy and Pharmaceutical Technology, University of Sevilla,* ***2.*** *Servicio de Medicina Nuclear. Hospital Universitario Virgen del Rocío,* ***3.*** *IBA Molecular*

Introduction: Crossing the BBB is a major challenge for the efficient delivery in the brain. Fortunately, exhibits transport mechanisms and there are numerous strategies to cross it. Transferrin (Tf) has been widely applied to enhance cellular uptake of nanoparticles (NPs) in brain delivery.

To evaluate the biodistribution of nanocarriers, NPs was radiolabeled with radioisotopes for evaluation in organs or tissues after administration.

Methods: NPs were prepared by emulsion solvent evaporation technique. The size and zeta potential (ZP) was measured and the morphology and shape were examined by using transmission electron microscope. The encapsulation efficiency were determined indirectly by HPLC.

After, Tf was conjugated on the surface of NPs using a carbodiimide method with EDC and NHS. Conjugation efficiency was measured indirectly.

For radiolabeled, lyophilized NPs were dispersed in NaCl in vials under vacuum. SnCl2-2-hydrate were dissolved in HCl 37% and was added with ≈74 MBq of 99mTc. Finally, the system was incubated for 10 min.

99mTc -NPs suspensions were analyzed by thin layer chromatography (TLC) with silica gel strips. With 0.9% NaCl as the mobile phase, free pertechnetate ran with the front, meanwhile particles stayed in the start. Using a solution of pyridine:acetic acid:water (3:5:15), radiocolloids remained at start, NPs migrated with Rf = 0.6- 0.8.

Results: The particle size, polydispersity index (PdI) and ZP of NPs were characterized by DSC. NPs had a mean size of 253.02±13.10 nm with a zeta potential of -3.30±1.74 mV. Their PdI of 0.043±0.03. After conjugation, NPs-Tf showed a mean size of 312.94±27.93 nm with a PdI less than 0.2 and a ZP of -2.40±0.56mV. The NPs had an average encapsulation efficiency of up to 65.5±2.65% and the conjugation efficiency was 60.4±3.77. T.E.M. analysis revealed that NPs had a solid structure and spherical shape.

PLGA nanoparticles with Tf and without surface modification were labeled with 99mTc with a yield ≥ 90 % (TLC). After radiolabeling, particles increased in size, ≈ 380 nm.

Discussion: NPs exhibits a suitable size, a high homogeneity, encapsulation and conjugation efficiency.

Radiolabeling process were reproducible and nanosystem is suitable for in vivo theragnosis.

Capsaicin-Coated silver nanoparticles inhibit amyloid fibril formation of serum albumin

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 197

Mr. Bibin G Anand [1], ***Dr. Karunakar Kar*** [1], ***Mrs. Kriti Dubey*** [1], ***Mr. Dolat Singh shekhawat*** [1]

1. *Indian Institute of technology, Jodhpur*

Here, we have synthesized capsaicin-coated silver nanoparticles (AgNPsCap) and have tested their anti-amyloid activity, considering serum albumin (BSA) as a model protein. We found that amyloid formation of BSA was strongly suppressed in the presence of AgNPsCap nanoparticles. However, isolated capsaicin and uncapped control nanoparticles did not show such inhibition effect. Bioinformatics analysis reveals CH-□ and H-bonding interactions between capsaicin and BSA in the formation of protein-ligand complex. These results suggest the significance of surface functionalization of nanoparticles with capsaicin which probably enables capsaicin to effectively interact with the key residues of the amyloidogenic core of BSA.

Diamond micro-cantilevers as transducers for Olfactory Receptors – based biosensors

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 238

Dr. Raafa Manai [1], Dr. Dounia Kamouni-Belghiti [1], Dr. Marie-Annick Persuy [2], Ms. Massiel Habchi [1], Dr. Lionel Rousseau [3], Ms. Maira Possas Abreu [3], Dr. Philippe Bergonzo [1], Dr. Edith Pajot-Augy [2], Dr. Guenhaël Sanz [2], Dr. Emmanuel Scorsone [1]

1. *CEA,* ***2.*** *INRA,* ***3.*** *ESIEE*

Introduction

Olfactory receptors (ORs) are proteins located in the plasma membrane of olfactory sensory neurons. They belong to the family of G protein-coupled receptors. Individual ORs bind various odorants with distinct affinities and specificities. Therefore, ORs combined into arrays are considered as highly promising for the conception of biosensors for odorant detection. Here we investigated a new approach consisting of chemically grafting the ORs onto synthetic diamond grown by Chemical Vapor Deposition. Because of its stability, biocompatibility, and ability to immobilize biological targets, diamond is a material of choice for biosensors.

Methods

ORs were grafted onto diamond surfaces using two routes, either (1) through covalent bonding of hexanoic acid radical on diamond followed by EDC/NHS peptidic coupling to an OR, or (2) using covalent attachment of nitriloacetic acid (NTA) on diamond as chelating agent, which binds an 6His-tagged OR via NTA-Ni interaction. Both grafting procedures were validated by electrochemical impedance spectroscopy (EIS) on boron doped diamond electrodes. They were then applied to bulk diamond micro-cantilevers freshly hydrogenated in hydrogen plasma in a Chemical Vapour Deposition reactor. Functionality for odorant detection was assessed in the liquid phase using a Laser Doppler read-out system.

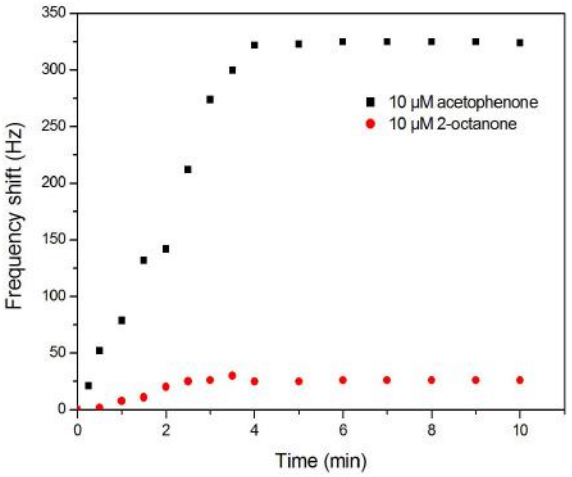
Results

OR7D4 was grafted through route (1), while 6His-tagged M71 was grafted through route (2). The Nyquist plots obtained from both EIS spectra exhibit an increase of the charge-transfer resistance, thus of the electrode coverage, showing for the first time an efficient immobilization of ORs on diamond.

Then, the 6His-M71 micro-cantilever sensor exhibited a good sensitivity to its known odorant ligand acetophenone, with a frequency shift near 100Hz for 1µM exposure (320Hz for 10µM), and a good selectivity against a non-ligand odorant. Likewise, the OR7D4 micro-cantilever sensor showed a sensitivity of 200Hz for exposure to its ligand androstenone at 1µM with a good selectivity against non-ligand odorants.

Discussion

Micro-cantilevers grafted with ORs are thus functional, and yield a quantitative, reproducible response to target analytes, at least at the micromolar level, and selectivity among odorants. The results are promising for the development of bioelectronic noses consisting of arrays of OR-grafted diamond-based resonators for specifically detecting target odorants.



Typical response of 6His-M71 based micro-cantilever

Figure iconan.jpg

Diphtheria toxin-derived protein for targeted delivery of nanoparticles

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 243

Mr. Mahesh Agarwal [1]***, Dr. Amaresh Sahoo*** [2]***, Dr. Biplab Bose*** [1]

1. *Indian Institute of technology, Guwahati,* ***2.*** *Indian Institute of Information technology, Allahabad*

Over the last one decade, various types of nano-formulations have been developed and approved for use. However, cell-specific delivery of nanoparticles (NPs) is still a challenge. Antibodies and peptides are often used for targeted delivery of NPs. In the present work, we have used a small protein derived from Diphtheria toxin to home NPs to specific cells. Diphtheria toxin (DT) binds to a specific receptor on cell surface and is internalized by receptor-mediated endocytosis. This receptor is overexpressed in various types of cancer cells. We have expressed the recombinant receptor-binding domain of DT (RDT) that binds to the cell surface receptor. We have coated PLGA NPs with RDT. Coating, with RDT, is revealed by UV spectroscopy and Bradford's assay. RDT coated NPs showed enhanced cellular uptake of NPs in U-87 MG cells with specific DT receptor. The internalization of RDT coated NPs is shown by Flow cytometer and Fluorescent microscope. These RDT-coated NPs can be used for delivery of a chemotherapeutic agent to cells expressing DT-receptors.

Formulation and In-Vitro Evaluation of Enteric Coated Capsules Filled with Recombinant Human Keratinocyte Growth Factor Loaded Chitosan Nanoparticles

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 245

Dr. Palanirajan Vijayaraj Kumar [1], ***Dr. Yeong Siew Wei*** [1], ***Mr. Mhd Luay Takahje*** [1], ***Dr. Lee Ming Tatt*** [1], ***Prof. Abu Bakar Bin Abdul Majeed*** [2], ***Dr. Vasudevan Mani*** [2]

1. *UCSI University, Faculty of Pharmacy, No. 1, Jalan Menara Gading, Taman Connaught, Cheras 56000, Kuala Lumpur,* ***2.*** *Universiti Teknologi MARA, Faculty of Pharmacy, Research Management Institute, Shah Alam*

Recombinant human keratinocyte growth factor (rHuKGF) is a protein used to treat oral mucositis, small intestinal ulceration and colitis in hematological malignancies patients who are receiving radio and chemotherapy. Mucoadhesive chitosan nanoparticles are used to develop oral delivery systems. In this project, rHuKGF loaded chitosan nanoparticles were prepared by ionotropic gelation of chitosan with tripolyphosphate anions with the presence and absence of PEG 2000 as a cryo-protectant. Size distribution and zeta potential of rHuKGF loaded chitosan nanoparticles were determined by using zeta-sizer. In addition, the morphology was determined by using field emission scanning electron microscope. Enzyme linked immunosorbent assay was used to determine the amount of rHuKGF that was loaded in chitosan nanoparticles. The prepared chitosan nanoparticles were filled in capsules and coated by enteric coating polymer kollicoat MAE 100 P, in-vitro dissolution studies were performed to evaluate the release of rHuKGF in a buffer solution having pH of the small intestine by using USP dissolution apparatus I. The rHuKGF loaded chitosan nanoparticles with and without PEG 2000 had spherical shape and positive surface charge. Particle size distribution of rHuKGF loaded chitosan nanoparticles was ranged from 119 to 385.7 nm. The rHuKGF loaded chitosan nanoparticles prepared with PEG 2000 capable to re-disperse fast without forming aggregates in simulated intestinal fluid pH 5.5 phosphate buffer solution. In-vitro release study showed the ability of kolicoat MAE 100 P to prevent the release of rHuKGF in simulated gastric fluid pH 1.2 and allow it to release in simulated phosphate buffer solutions pH higher than 5.5.

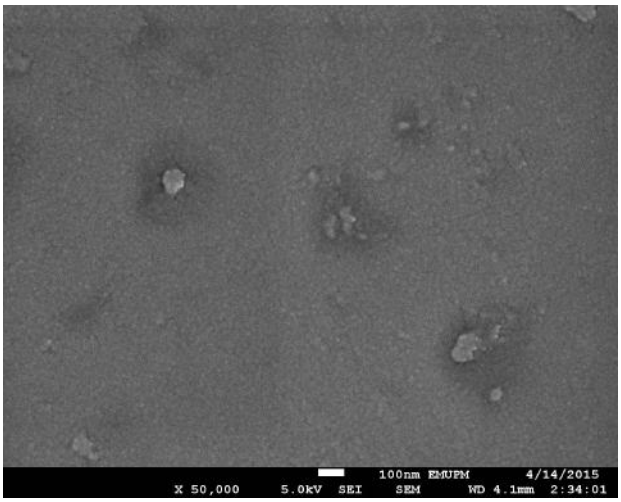


Image shows the scanning electron micrograph of rhukgf loaded chitosan mucoadhesive nanoparticles with peg 2000.jpg

Molecular Imprints for the Detection of Specific Glycoproteins Implicated in Cancer

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 285

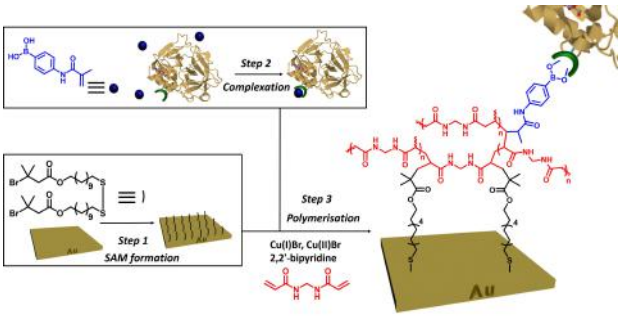
Ms. Philippa Mitchell [1], ***Dr. Lewis Hart*** [1], ***Prof. Paula Mendes*** [1]

1. *University of Birmingham*

Molecular diagnostics rely heavily on immuno-detection, such as the enzyme-linked immunosorbent assay (ELISA).1 One of the key drawbacks of employing antibodies in biomedical detection systems is their inability to differentiate between different glycoforms of the same protein.2 3 One such ELISA, where this short-coming is pronounced is the prostate specific antigen (PSA) ELISA developed for the diagnosis prostate cancer.4 There are multiple glycoforms of PSA including those that are strongly suggested as biomarkers of prostate cancer, however the ELISA, which is considered the 'gold standard, cannot distinguish between normal and cancerous glycoforms making the test uninformative, unreliable and inaccurate.5 6 To this end, we look to utilise molecular imprinted polymers (MIPs) to produce a novel diagnostic method for the detection of cancerous PSA glycoforms.

MIPs within ultra-thin hydrogels are synthesised from a surface using controlled living polymerisation (Figure 1). An initiator molecule (11'-dithiobis[1-(2-bromo-2-methylpropionyloxy)undecane]) is synthesised and incubated with gold to form a self-assembled monolayer (SAM). In a separate step, PSA is incubated with vinylboronic acid to form a complex. The complex is then incubated with the initiator surface and polymerisation triggered around the complex using N,N'-methylenebisacrylamide (MEBA), Cu(I)Br and 2,2'-bipyridine as the metal-ligand catalyst and ethyl-2-bromoisobutyrate as a sacrificial initiator to form an imprint in biologically compatible conditions.

The afforded SAMs were in good agreement with the literature, measuring 1.74 ± 0.12 nm in thickness and with a water advancing angle of 70 ± 1.8°, as evaluated by ellipsometry and contact angle, respectively.7 The extent of the polymer network was determined by the ratio of Cu(I)Br to Cu(II)Br. The resultant system was then optimised to produce MEBA layers with a thickness of 8.07 ± 0.76 nm, which will act as the foundation into which PSA imprints will be formed. We now look to incorporate boronic acid monomers complexed with PSA into the polymerisation network to create MIPs capable of differentiating between the subtle differences in the glycosylation characteristic of each PSA glycoform, to thus produce an informative, reliable and accurate test for prostate cancer.



Mip for paris.jpg

Optimizing liposomes delivering arsenic trioxide to cervical cancer cells in vitro

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 317

Ms. Anam Akhtar [1], ***Dr. Xuesong Wen*** [1], ***Dr. Scarlet Wang*** [1], ***Dr. Lucy Ghali*** [1], ***Dr. Celia Bell*** [1]

1. *Middlesex University*

Arsenic trioxide (ATO) has been demonstrated to have significant therapeutic effects against acute promyelocytic leukemia and various haematological malignancies. However the lack of success in clinical trials in treating solid tumours due to its toxicity has prevented its further application. In this study, ATO encapsulated liposomes were prepared and investigated for their physico-chemical characteristics, drug loading efficiency and inhibitory activity on cervical cancer cells under different surface charges (neural, negative or positive charges) and sizes (100, 200 and 400nm). Five different liposomal formulations were prepared by conventional lipid film hydration technique. Liposomes of increasing sizes were prepared by extrusion from filters of 100nm, 200nm and 400nm pore sizes respectively. Charged liposomes were prepared in a similar way by incorporating dimethyldioctadecylammonium bromide (DB) and 1-stearoyl-2-hydroxy-sn-glycero-3-phospho-sodium salt (DSPG) as cationic and anionic charge carrier lipids for liposomes. The resultant liposomes were stable at 4°C with less than 10% arsenic leakage after a month as determined from inductively coupled plasma optical emission spectroscopy (ICP-OES). Loading efficiency was observed to be independent of tested size range; however neutral liposomes had the highest loading efficiency among the charged liposomes. Cellular uptake and apoptosis studies of these liposomes were evaluated against HPV positive (HeLa) and HPV negative (C33a) cervical cancers and a normal control cell line (CRL 1790) through inductively coupled plasma-mass spectroscopy (ICP-MS), flow cytometry and toxicity studies. Liposomal uptake was found to be independent of sizes but dependent on surface charges and cell lines. Positive liposomes displayed the highest uptake, but the highest toxicity to the cells after their exposure rules them out as suitable candidates as drug carriers. Lower toxicity to the cells was observed when liposomal ATO was used instead of free drug and overall, C33a cells were more susceptible to ATO than HeLa despite a lower arsenic uptake. Cancer cells displayed the significant apoptotic effect and toxicity per unit uptake of ATO as opposed to normal cells when liposomal ATO was employed. In conclusion, neutral 100 nm liposomes were chosen as an effective ATO carrier to cervical cancer cells due to their lower toxicity, higher loading efficiency and high stability.

Study Effects of Synchronize Utilize of Gamma-ray and Silver Nano Particles Treatment on Human Prostate Cancer Cells

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 329

Dr. alireza shams [1]

1. *Alborz University of Medical Sciences, Karaj, Iran*

Background: Prostate cancer is the second most common cancer in America which causing great harm and waste cost. Furthermore more prostate cancer treatment is ineffective. Purpose of this study was to evaluate the effects of therapy and the rate of increase of the absorbed dose of gamma radiation with silver nano particles in treatment of prostate cancer.

Materials & Methods: DU145 cell line originating from Human prostate cancer was purchased from Pasteur Institute. After thawing of defreezed samples, cells were incubated with DMEM medium and 15% FBS serum was added. For preventing contamination amphotericin B in 25 µM/ml was added. Cells were incubated over a period of 3 to 5 days to reach a good Con-fluency then cells divided into 4 groups. Tentatively assigned to the control group, the second experimental group treated with gamma irradiation at a doses of 2, 6 and 10 Gray(Gy), the third group treated with 53 µ g / ml silver nano particles, The fourth group includes simultaneous treatment with gamma doses and silver nano particle. Cell groups studied by staining with trypan blue as well as by MTT assay (ELISA) reader.

Results: The results showed the use of gamma rays and silver nano particles caused a significant reduction in the number of cancer cells in the treated groups compared to the other treatment groups and a control group. Using of silver nano particles as a radio sensitize and radiation therapy in prostate cancer cell lines DU145 resulted in the increase of the gamma-ray photon energies of 6 and 10 Gy

Discussion: The gamma photon radiation to the tumor cells were incubated nano particle probability Photo-electric process as a process when handling the extremely high energy gamma photons hitting the silver nano particles to cancer cells, photo electron and Auger electrons (Auger Electrons) and secondary electrons produced by secondary particles can be It will not be very effective, leading to failures in the field of DNA damage and cell death.

Keywords: Prostate cancer, gamma - ray , Silver nano particles, DU145, Cell Culture

RADIOLABELED NANOPARTICLES STRATEGIES FOR BREAST CANCER THERAGNOSIS

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 458

Dr. Lucía Martín-Banderas [1], ***Ms. ROCÍO IGLESIAS JEREZ*** [2], ***Mrs. Mª Dolores Cayero-Otero*** [3], ***Dr. Alejandro Perera*** [4], ***Mr. Roque Salazar Cabrera*** [5], ***Ms. Isabel Borrego Dorado*** [2]

1. *Pharmacy and Pharmaceutical Technology, University of Sevilla,* ***2.*** *Servicio de Medicina Nuclear. Hospital Universitario Virgen del Rocío,* ***3.*** *Universidad de Sevilla,* ***4.*** *Centro de Isotopos (CENTIS). La Habana,* ***5.*** *IBA Molecular*

Introduction

Nanomedicine offers an exciting new paradigm change for real medicine, which can be translated into a huge impact on health care. This is mainly due to the possibility of combination treatment strategies and diagnostic techniques, known as theragnosis.

Objective

The objective of present work is to develop double functionalized PLGA nanoparticles for breast cancer theragnosis. PLGA nanoparticles were radiolabeled with 99mTc and particle surface were modified with a monoclonal antibody (MAb) against HER2+ cancer cells.

Materials and Methods

PLGA nanoparticles were prepared by nanoprecipitation method [1]. Loaded nanoparticles were prepared dissolving paclitaxel (a gift from PhytoBiotech, Canada) into polymer solution in acetone at 10 % w/w concentration.

Different radiolabeling strategies were carried out [2]:

(i) PLGA nanoparticles were surface modified with a MAb against HER2 + cancer cells using the carbodiimide strategy. Then, nanoparticles were radiolabeled with 99mTc directly using SnCl2.

(ii) Antibody was firstly reduced using a ß-mercaptoethanol. For this purpose, MAb was incubated for 30 min at room temperature. After purification (Amicon 30K) to eliminate the excess of ß-mercaptoethanol, the reduced antibody was labeled with 99mTc (10 mCi) via Sn2+ reduction of pertechnetate, using sodium pirophospate (Angiocis® 20mg) as a weak competing ligand.

MAb Conjugation Efficiency was evaluated by Bradford method.

Results and Discussion

We obtained paclitaxel-loaded PLGA nanoparticles 190 nm in diameter with a zeta potential around -20 mV. Encapsulation efficiency was 85% (determined by HPLC). After antibody conjugation particles diameter increases slightly up to 250 nm in diameter. ZP values were nearly neutrality which indicates the presence of MAb on particle surface. Antibody conjugation efficiency was around 90%. That means 30-35 □g of MAb per □g□of nanoparticle.

Both radiolabeling strategies were highly effective (up to 98%).

References

[1] Fessi, H., Puisieux, F., Devissaguet, J.P., Ammoury, N., Benita, S. Int. J. Pharm. (1989) 55, R1-R4

[2] Psimadas D, Bouziotis P, Georgoulias P, Valotassiou V, Tsotakos T, Loudos G. Contrast Media Mol Imaging. (2013), 8(4):333-339

Acknowledgements
Authors are special grateful for the financial support from Fundación Progreso y Salud (Junta de Andalucía) and Janssen Pharmaceuticals. Authors also thanks PhytoBiotech (Canada) for paclitaxel supply

Mesoporous silica nanoparticles surrounded by a lipid bilayer: Absence of in vitro hemonanotoxicity

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 369

Dr. Christina Grigorakaki [1], ***Dr. Vincent Schlesser*** [2], ***Dr. Céline Hoffmann*** [1], ***Dr. Jean-Sébastien Thomann*** [3], ***Ms. Gaëlle Corne*** [3], ***Dr. César Pascual Garcia*** [3], ***Dr. Sivashankar Krishnamoorthy*** [3], ***Prof. Rolf Bjerkvig*** [1], ***Dr. Valérie Palissot*** [1]

1. *Luxembourg Institute of Health,* ***2.*** *Centre Hospitalier de Luxembourg,* ***3.*** *Luxembourg Institute of Science and Technology*

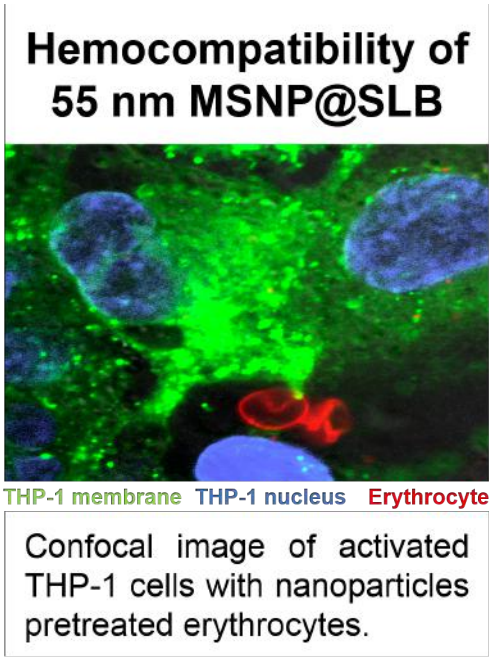
Nanomedicine aims at rising innovative tools in order to improve the diagnosis and the therapy of human diseases. The development of robust theranostic nanomaterials displaying treatment efficiency and reduction of detrimental side effect to normal tissues will deeply improve patient quality of life. Due to their size and versatile character nanoparticles can pass through the physiological barriers and deliver medicines. Therefore, the translation process of these new therapeutic nanocarriers to their clinical use relies on their interaction with the physiological environment.

Following the synthesis and the complete physicochemical characterization of a 55 nm mesoporous silica nanoparticle surrounded by a lipid bilayer (55 nm MSNP@SLB), we have evaluated its susceptibility for safely intravenous administration. In a first step of in vitro experimentation on red blood cells (RBCs), Scanning Electron Microscopy and hemolysis assay have shown no deformation and disruption of their membrane. No cytotoxic effect was neither observed on Peripheral Blood Mononuclear and on Human Marrow Stromal cells (HS-5 cell line).

We have therefore elaborated our nanoparticle study with the determination of coagulation parameters i.e. Prothrombin Time (PT) and Activated Partial Thromboplastin Time (APTT). These Nanoparticles display values in a normal range without formation of a clot. These results are in correlation with no platelets activation and aggregation.

Using scanning confocal microscopy, we observed that isolated RBCs that have been in contact with nanoparticles surrounded by a lipid bilayer are not cleared by activated macrophages (THP-1). This phenomenon is correlated with the absence of phosphatidylserine externalization on RBCs membrane. The activated macrophage model incubated with the nanoparticles of interest showed uptake of nanoparticles and did not trigger ROS production.

In conclusion, the nanoparticle under development displays no in vitro toxic effects in the major cellular components of blood and the development of this multiphased nanoplatform should result in a biocompatible theranostic tool.



Iconan 2016 png grigorakaki.png

Novel Synthetic Method and Characterization of Magnetic Resonance/Near-infrared dual-modal imaging agents by In Vivo Molecular Imaging

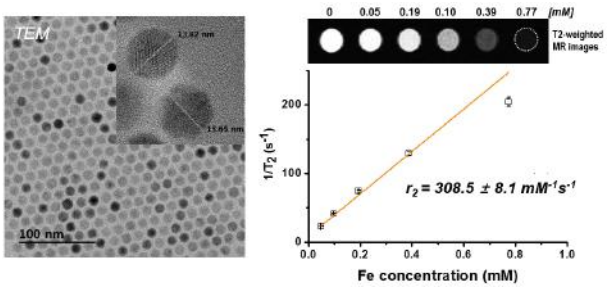
Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 372

Mr. Hyunseung Lee [1], ***Ms. Hyun Min Kim*** [1], ***Dr. Jee-Hyun Cho*** [1], ***Mr. Jongeun Kang*** [1], ***Dr. Hye Sun Park*** [1], ***Dr. Kwan Soo Hong*** [1]

1. *Bioimaging Research Team, Korea Basic Science Institute*

A novel magnetic resonance (MR)/near-infrared (NIR) imaging contrast agent was developed by a facile fluorescent dye-coating on magnetic nanoparticles (MNP). The fluorescent dye molecules were directly attached onto the nanoparticle surface, which enable the nanoparticles to transfer from hydrophobic phase to hydrophilic phase. This facile synthesis method was applied to various fluorescent dyes and another hydrophobic nanoparticle to evaluate the synthetic principle. And the dye coating on the particle was well characterized by single particle imaging technique. We also demonstrated that photostability of fluorescent dyes on the particle surface were improved compared to the soluble dye form by monitoring of fluorescence signals under the continuous irradiation.

The dye-coated MNP presents high T2 relaxivity value (308 mM-1s-1), good cell viability due to biocompatible NIR dye, indocyanine green, supporting non-invasive in vivo MR/NIR imaging. We performed in vivo tracking of the dye-coated MNP labeled dendritic cells (DCs). Importantly, the migration of DCs via lymphatic drainage and homing into the lymph node were monitored through real-time NIR fluorescence and MR imaging. This novel MR/NIR contrast agent has potential applications as a non-invasive imaging agent for imaging or tracking of immune cells to confirm immunotherapeutic efficacy.



Abstract figure.jpg

3D Printing Revolution for Nanotechnology and Nanomedicine

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 382

Dr. Alessandro Paolini [1], Ms. Antonella Celluzzi [1], Dr. Antonella Baldassarre [1], Dr. Simona Sennato [2], Dr. Francesco Mura [3], Dr. Federico Bordi [2], Dr. Alberto Eugenio Tozzi [1], Dr. Andrea Masotti [1]

1. *Ospedale Pediatrico Bambino Gesù,* ***2.*** *CNR-ISC UOS Roma, c/o Dipartimento di Fisica, Sapienza Università di Roma,* ***3.*** *CNIS, Università di Roma Sapienza*

Introduction

The 3D printing is about to really transform our lives. While traditional laser and inkjet printers only make marks on paper, 3D printers build up solid objects one very thin layer over another. We are beginning to be surrounded by many prototypes, jewelry, sunglasses, works of art, toys and vehicle parts, but the next expected revolution is going to impact on the nanotechnology and biomedical fields.

Methods

Here we show how the 3D prototyping of a cell culture device gives the opportunity to easily culture cells on a sheet of carbon nanotubes, referred as “buckypaper”, and that these cells can be easily transduced with microRNAs, small RNA molecules able to regulate gene expression post-transcriptionally.

We printed 3D scaffolds using a commercial home-made printer (3DRag) and we studied polymer-coated buckypapers (BP) supported by them. We also assessed their toxicity compared to uncoated BP on Hek cells.

Results

3D scaffolds enable to obtain a versatile support to study BP toxicity and cell culture adhesion and proliferation. Polymer coated BPs have been demonstrated to be less toxic than the uncoated BP.

Discussion

Interestingly, buckypaper is only one of the numerous examples of biomaterials that it is possible to employ in biomedical applications for culturing cells. Therefore, we envisage that in the near future a growing number of materials and potential applications in the biomedical filed will appear on the horizon. 3D printed devices can open new perspectives not only for targeted manufacturing in the biomedical field but also for producing novel materials and ‘bio-devices’ for nanomedicine applications.

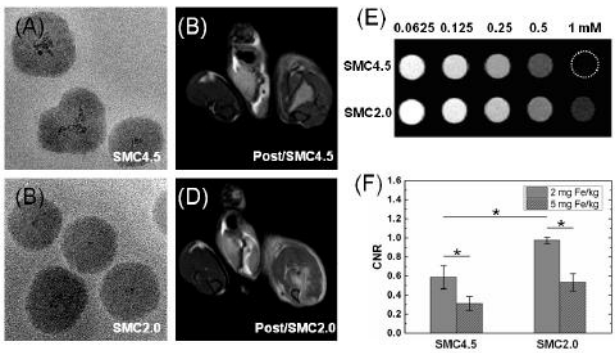
Synthesis of High Sensitive Magnetic Nanoparticles and Application for Inflammation Detection

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 399

Mr. Jongeun Kang [1], ***Mr. Hyunseung Lee*** [1], ***Dr. Kwan Soo Hong*** [1]

1. *Bioimaging Research Team, Korea Basic Science Institute*

Magnetic nanoparticles (MNPs) have been widely used as contrast agents for magnetic resonance imaging (MRI), drug delivery, and hyperthermia agents. Among them, MRI is one of the important applications of MNPs because the low sensitivity of MRI can be improved by MNPs. The improvement of T2 relaxivity of MNPs still remains as a critical issue. In this study, we reported an easy and large scale-up synthesis technique for high sensitive MNPs via pH regulation during co-precipitation-based method. A large amount of the aggregated MNPs by regulating pH were successfully coated with silica, in which the number of MNP cores in the silica was simply controlled by pH. As a result, the MNPs could form a nanocluster by decreasing repulsion force for each other when the particle surface charge is almost zero in neutral pH state. We report two types of silica-coated magnetic nanoclusters (SMCs) synthesized at pH2.0 and pH4.5, named as SMC2.0 and SMC4.5, respectively. SMC4.5 provides improved T2 relaxivity (270 mM-1s-1 vs. 155 mM-1s-1 for SMC2.0) due to increased number of cores (18 and 6 for SMC4.5 and SMC2.0, respectively). For in vivo T2*-weighted MRI experiments of inflammation animal model, SMC4.5 provided more specific and high sensitive detection of inflammation following in vivo labeling of the SMC4.5 in macrophages in the sites, showing a possibility of minimum dosage due to higher sensitivity. The nanocluster-type SMCs could be a T2 MRI contrast agent with minimum side-effect disease diagnosis and bio-imaging applications.



.png

Use of cell membrane permeating peptides for delivery of plasmid DNA into dendritic cells

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 415

Ms. Dominika Hobernik [1], ***Dr. A. James Mason*** [2], ***Prof. Stephan Grabbe*** [1], ***Dr. Matthias Bros*** [1]

1. *Department of Dermatology, University Medical Center of the Johannes Gutenberg University Mainz,* ***2.*** *Institute of Pharmaceutical Science, King's College London*

Immune therapy is a promising approach in the treatment of cancer. Besides ex vivo incubation of dendritic cells (DC) with tumor-associated antigens (TAA) and adjuvants, an increasing number of nanosized systems that co-deliver these compounds are assessed for effective in vivo drug delivery. DNA vaccines that encode for TAAs are an inexpensive, stable and safe alternative to protein- and peptide-based vaccines. Still, efficient delivery of pDNA in vivo is a challenge that needs to be faced.

LAH4-L1 is a cell membrane permeating peptide shown to complex pDNA and RNA. We analysed the suitability of this peptide to deliver pDNA to DC. First, we incubated murine spleen cells with fluorescently labelled LAH4-L1/pDNA complexes and analysed transfer of fluorescently labelled pDNA to different immune cell subtypes via flow cytometry. Due to strong interaction with endocytically active macrophages and DC, we also analysed bone marrow-derived dendritic cells (BMDC). BMDC showed strong binding/internalization of LAH4-L1 complexed pDNA. Unexpectedly, these BMDC also showed strong upregulation of surface markers that indicate cellular activation (MHCII, CD40, CD80, and CD86). In agreement, LAH4-L1/pDNA-treated BMDC generated strongly enhanced levels of proinflammatory cytokines (IL-1ß, TNF-a, IL-6). These findings suggest that LAH4-L1 may exert a potent stimulatory capacity on DC which is crucial for the induction of an adaptive immune response. LAH4-L1 mediated stronger transfection efficiency than JetPEI, often used for the transfection of DC-like cell lines with transgene-encoding pDNA, as assessed using a luciferase reporter. Ongoing work is focussed on the use of LAH4-L1 for transfection of DC populations with antigen-encoding mRNA and pDNA and subsequent DC/T cell co-cultures to assess their T cell stimulatory capacity.

Future work is focussed on functionalization of LAH4-L1 with targeting moieties to target DC specifically. Furthermore, TAA-encoding pDNA will be used for in vivo proliferation assays and to induce tumor-specific immune responses.

Efficient gene silencing with siRNA formulated in DOP-DETA-based lipid nanoparticles

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 420

Dr. Tomohiro Asai [1], ***Ms. Mariko Sako*** [1], ***Ms. Jiao Li*** [1], ***Ms. Ayaka Okamoto*** [1], ***Dr. Takehisa Dewa*** [2], ***Prof. Naoto Oku*** [1]

1. *University of Shizuoka School of Pharmaceutical Sciences,* ***2.*** *Nagoya Institute of Technology*

Development of small interfering RNA (siRNA) therapeutics is a promising approach to address unmet medical needs. However, because RNA is easily degraded by RNases and hardly internalized into the cells, a suitable delivery system is required to develop siRNA therapeutics. Development of a siRNA delivery vehicle is the most urgent and crucial thing for clinical success of siRNA-based therapies. In the present study, we designed and synthesized a novel polycationic lipid derivative, a dioleoylphosphate-diethylenetriamine conjugate (DOP-DETA) for the purpose of efficient siRNA delivery. Polycationic liposomes composed of DOP-DETA, cholesterol, and dipalmitoylphosphatidylcholine (DPPC) were prepared and mixed with siRNA to obtain lipid nanoparticles (LNP) loaded with siRNA. To evaluate the delivery potential of the LNP, a reporter gene silencing assay was performed using human fibrosarcoma cell line HT1080 constitutively expressing green fluorescent protein (GFP). The data showed that the LNP loaded with siRNA for GFP induced gene silencing at the concentration of less than 5 nM of siRNA. The gene silencing efficiency and cellular uptake amount of siRNA were dependent on siRNA/lipid molar ratio and increased with increasing concentration of lipids. Confocal microscopic observation demonstrates that siRNA transfected with the LNP diffusely distributed throughout the cytoplasm of the cells. Next, the LNP loaded with siRNA for poly [ADP-ribose] polymerase 1 (PARP1-siRNA) was prepared to evaluate its potential to suppress the expression of the innate protein. The results showed that knockdown of PARP1 protein was markedly observed when murine Lewis lung carcinoma cells had been transfected with 0.5 nM of PARP1-siRNA. The silencing efficiency of the LNP loaded with PARP1-siRNA was approximately 80-fold higher than that obtained with the common transfection reagents. The present findings suggest that DOP-DETA-based LNP might have excellent potential to deliver siRNA to target cells.

Glimepiride solid lipid nanoparticles: Formulation, Evaluation and In-Vivo study.

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 430

Dr. omneya khowessah [1]

1. *Faculty of pharmacy, Cairo university*

Introduction: Solid lipid nanoparticles(SLNs) have been reported as an alternative drug delivery system to traditional polymeric nanoparticles. Many studies were carried out and aimed to determine the enhancement ability of (SLNs) on the solubility of many poorly water soluble drugs and consequently, the improvement of their bioavailability.

Methodolgy: To enhance the bioavailability of glimepiride, SLNs were formulated using two different polymers (compritol and glycelymonostearate) in a fixed drug: lipid polymer ratio (1:20). Two different ratios of three different surfactants were used. GMP-SLNs were prepared adapting two different techniques namely, hot fusion method and solvent emulsification method. The prepared GMP-SLNs were evaluated with respect to encapsulation efficiency, yield value ,particle size, zeta potential, image analyzer, and TEM analyzer. The DSC analysis and PXRD analysis were carried out on selected formulae. The pharmacokinetic study of the selected GMP-SLNs formula (F2) was carried out on male Newzeland white rabbits and the results were compared with those of marketed product (Amaryl ® 4mg tablets).

Results: The resulted values revealed that most of the solid lipid nanoparticles were of circular shape, the particle size ranging from 104 nm to 334 nm with zeta potential ranged from -9.34 mV to -38.6 mV. The resulted EE% values ranged from 97.8±0.25% to 99.7± 0.23%. The yield values were from 97.88±2.37 % to 98.96±2.03%. TEM analysis of SLNs formulae F2 and F8 showed rounded and discrete shaped particles. The DSC analysis results of F2 and F8 showed that there were no interactions between the drug and the used excipients. PXRD diffractograms of F2 and F8 showed lamellar lattice in case of SLNs and crystalline lattice in case of the compritol, glyceryl monostearate and drug powders in bulk state. The pharmacokinetic study and statistical analysis of the selected formulae (F2) showed that (F2) significantly enhanced the rate of GMP absorption from GIT and at 0.5, 2 & 6 hrs compared to the marketed product. The resulted Cmax was 28.1mg/ml, Tmax was 5.94 hrs, t1/2 abs was 1.53 hrs, t1/2ele. was 18.1 hrs, (maximum amount of drug absorbed) was 2018 µg, F was 1 and the bioavailability (F relative) was 100%.

	(%±S.D)	Size (nm± S.D)		(mV± S.D)	(%Yield± S.D)
F1	98.8± 0.30	352.4 ± 22	0.453 ±0.061	-33.2 ±0.05	98.85±1.62
F2	99.4± 0.66	246.6 ±30	0.324 ±0.038	-37.5 ±0.07	99.19±1.54
F3	99.7± 0.23	465.8 ±17	0.658 ±0.023	-24.3 ±0.03	97.88±2.37
F4	99.3± 0.12	342.6 ±23	0.531 ±0.072	-28.8 ±0.04	98.96±2.03
F5	97.8± 0.25	875.4 ±42	0.757 ±0.033	-13.2 ±0.06	97.91± 1.63
F6	98.7± 0.42	552.3 ±27	0.302 ±0.026	-11.5 0.05	98.24±3.23
F7	99.5± 0.32	843.6 ±38	0.637 ±0.044	-27.4 ±0.06	98.74±1.07
F8	99.3± 0.81	622.4 ±26	0.462 ±0.037	-34.3 ±0.07	99.15±1.06
F9	99.6± 0.31	644.2 ±34	0.476 ±0.072	-22.2 ±0.08	98.87± 3.08
F10	99.3± 0.70	487.4 ±27	0.323 ±0.025	-25.1 ±0.09	98.31± 1.82
F11	99.7± 0.21	784.4 ±38	0.622 ±0.044	-12.4 ±0.06	98.65± 2.08
F12	99.5± 0.53	579.0 ±28	0.412 ±0.062	-10.2 ±0.08	98.32± 2.05

Table.png

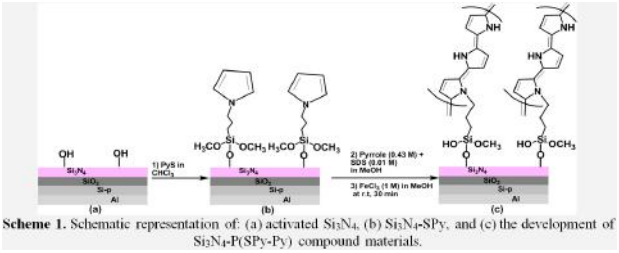
A novel immunosensor based on polypyrrole electroless deposition on silicon nitride substrates: Interleukine-10 detection

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 478

Ms. Faiza Nessark [1], Dr. Abdoullatif Baraket [2], Dr. Nadia Zine [3], Prof. Ahmed Zouaoui [4], Prof. Belkacem Nessark [5], Prof. Joan Bausells [6], Prof. abdelhamid Errachid [3]

1. *Laboratoire d'Electrochimie et Matériaux (LEM), Université Ferhat Abbas Sétif 1, 19000, Algérie,* ***2.*** *Université Claude Bernard Lyon1,* ***3.*** *Institut des sciences Analytiques (ISA) , Université Claude Bernard Lyon1,UMR 5280, 5 rue de la Doua,69100 Villeurbanne Lyon, 6 France,* ***4.*** *Laboratoire de Croissance et Caractérisation de Nouveaux Semi-conducteurs (LCCNS), Université Ferhat Abbas Sétif 1, 19000, Algeria,* ***5.*** *Laboratoire d'Electrochimie et Matériaux (LEM), Université Ferhat Abbas Sétif 1, 19000, Algeria,* ***6.*** *Instituto de Microelectronica de Barcelona, IMB-CNM (CSIC), Campus UAB, 08193, Bellaterra, Spain.*

We report in present study about development of a novel strategy of anti-human interleukin-10 (anti-IL-10) antibody immobilization onto polypyrrole (PPy) modified capacitance substrate. Here, we propose the development of a biosensor based on silicon nitride (Si3N4) chemically modified by a deposition of a PPy layer. In order to ensure adhesion of the conductive organic polymer to the substrate, a silane-pyrole reagent (SPy) N-(3-trimethoxysilylpropyl) pyrrole was used to create a covalent bridge between the silicon substrate and the layer of PPy (Scheme 1). The surface morphology of the composite Si3N4-P(SPy-Py) was studied by scanning electron microscopy (SEM) and infrared spectroscopy (FTIR). The electrochemical behavior of the novel composite Si3N4-P(SPy-Py) was characterized by cyclic voltammetry (CV) and electrical impedance spectroscopy (EIS). The (Si3N4)-P(SPy-Py) composite was subsequently electrochemically modified by diazonium salts (4-Aminophenylacetic acid) (CMA) for the immobilization of specific antibodies (anti-human interleukin-10 monoclonal antibody) by carbodiimide chemistry. The configured immunosensor successfully detected human IL-10 in the range of 1-50 pg/mL. This new composite shows a high sensitivity and a novel approach for the development of new strategies for biosensor development and detection.



Scheme 1. Schematic representation of: (a) activated Si_3N_4, (b) Si_3N_4-SPy, and (c) the development of Si_3N_4-P(SPy-Py) compound materials.

Scheme 1.png

Biophysical Characterization, Nanoscale Composition and Cell Uptake Studies of pH-Sensitive Drug Delivery Systems

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 490

Dr. Jana B. Nieder [1], ***Ms. Rasa Ozolina*** [2], ***Ms. Ana M. Carvalho*** [3], ***Ms. Vânia Vilas-Boas*** [2], ***Prof. M.E.C.D. Real Oliveira*** [3], ***Dr. Marlene Lúcio*** [3]

1. *INL-International Iberian Nanotechnology Laboratory,* ***2.*** *INL - International Iberian Nanotechnology Laboratory,* ***3.*** *University of Minho*

Nanocarrier-based chemotherapy is one of the few nanotechnology-based medical therapies that reached the clinics. This happened already in 1995, when the commercial liposomal anti cancer drug delivery formulation DOXIL® was introduced in the market. Albeit these early developments, still today nanotechnology-based drug delivery systems are far from reaching optimal selectivity and controlled release ability.

In our study we use different liposomal formulations designed for pH-sensitive drug release and study their biophysical characteristics, when used for trafficking paclitaxel (PTX) and doxorubicin (DOX), both widely used chemotherapeutic anti-cancer drugs.

Our work describes a combined spectroscopy and imaging approach to evaluate the biophysical properties of liposomal formulations. We study the nanoscale composition of the nanocarriers using molecular rulers in a fluorescence quenching assay, and analyze the cell uptake characteristics based on the autofluorescence of DOX using confocal microscopy.

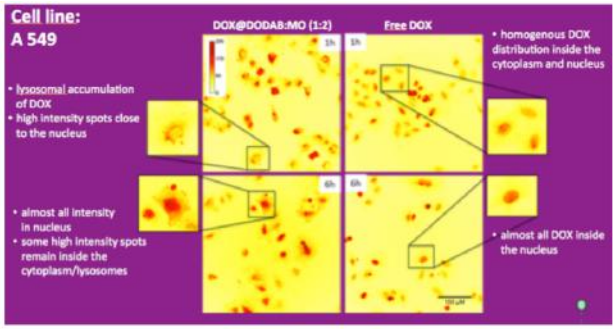


Figure: Cell uptake study: Confocal images taken on fixed cells incubated with liposomal formulations and free DOX.

Nieder-celluptakeiconan2016.png

Effect of Nanosilver with salicylic acid & nano salicylic acid pre-treatment on Antioxidant responses in Dracocephalum moldavica L.

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 513

Prof. Hossein Abbaspour [1], ***Dr. zahra haghighipak*** [1], ***Dr. Naser Karimi*** [2]

1. *Damghan Branch, Islamic Azad University,* ***2.*** *Razi University*

Nanoparticles (NPs) with different composition, size, and concentration have been reported to influence growth and development of various plant species with both positive and negative effects. Ag-nanoparticle is important for its characteristic physiochemical and biological properties. Dracocephalum moldavica L. is well known as medicinal plants because of its biological and pharmacological properties. Salicylic acid (SA) is an important signal molecule modulating plant response to stress. In here, we investigated some antioxidative responses of D. moldavica under Ag-NPs pretreated with different SA and nano-SA concentrations. The study was carried out in a randomized block design with 3 replications in four levels of SA and nano SA (0, 20, 60, 80 mM) and six Ag-NPs treatments (0, 10 ,20, 40, 60, 80 PPm) for 21 days. AgNPs significantly induced oxidative stress marker such as H2O2 content in a concentration dependent manner. The phytotoxicity of AgNPs led to an increase in catalase and peroxidase activities and synthesis of antioxidant compounds such as carotenoids, proline and total soluble carbohydrate. Further results showed that SA and nano-SA pretreatments ameliorates the Ag-phytotoxicity, which nano-SA was more effective than SA in this manner.

Formulation of Levofloxacin Loaded Niosomal suspensions as an Ocular Delivery System: In-Vitro Evaluation and Microbiological Study.

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 391

Dr. salwa hussien [1]

1. *Faculty of pharmacy, Cairo university*

INTRODUCTION

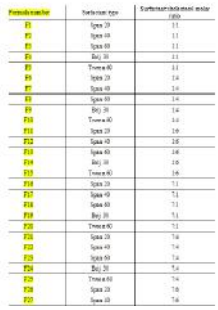
Levofloxacin hemihydrate is a fluoroqinolone antibiotic with broad spectrum antibacterial agent. Encapsulation of the drug into vesicular system provides delivering drug in a controlled manner to enhance bioavailability and get therapeutic effect over a longer period of time. Niosomes are such hydrated vesicular systems containing nonionic surfactants along with cholesterol or other lipids delivering drug to targeted site which are non toxic, requiring less production cost, stable over a long period of time in different conditions.

METHODS

Niosomes of levofloxacin hemihydrate were prepared by reverse evaporation method.Optimization of the formulation step was carried by application of 5x3x2 factorial design to study the influence of three parameters including surfactant type , cholesterol molar ratio and surfactant molar ratio on the physicochemical properties including particle size , entrapment effeciency (E.E) and release parameters (Q8h and Q24h). Transmission electron microscope (TEM) was carried out for the selected formula. Microbiological study of the selected formula was also investigated.

RESULTS AND DISCUSSION

Niosomes prepared from span 40 and span60 showed the highest entrapment efficiency. The best formula was the one which prepared from span60 and cholesterol with molar ratio(7:6) of entrapment efficiency 84.1% and particle size (587nm). In-vitro drug release results confirmed that niosomal formulations have exhibited a high retention of levofloxacin inside the vesicles and can sustain release up to 24 hours. The in-vitro evaluation of levofloxacin hemihydrate niosomes in comparison to levofloxacin hemihydrate solution showed the effect of niosomes in prolongation of drug release from the ocular delivery system. Niosomes may be considered as promising ophthalmic carriers for the topical application of levofloxacin hemihydrates .The antimicrobial activity of the selected formula was retained up to 24 hours



Niosomes table.png

Sickle cell hemoglobin detection in drying drops: from protein/nanoparticle interaction to low resource diagnostic tools

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 459

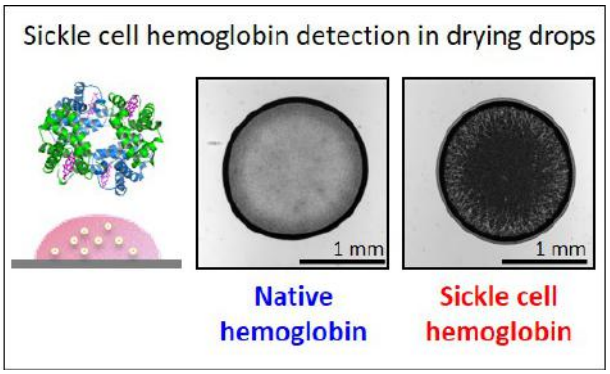
Dr. Stephanie Devineau [1], ***Dr. Manos Anyfantakis*** [2], ***Mr. Laurent Marichal*** [3], ***Dr. Laurent Kiger*** [4], ***Prof. Damien Baigl*** [2]

1. *Centre For BioNano Interactions (CBNI), University College Dublin,* ***2.*** *Department of Chemistry, Ecole Normale Superieure,* ***3.*** *CEA Saclay, NIMBE,* ***4.*** *Inserm, IMRB*

The coffee-ring effect denotes the accumulation of particles at the edge of an evaporating drop pinned on a substrate, as one can observe in the black ring of a coffee stain. Because it can be detected by simple visual inspection, this ubiquitous phenomenon has been recently combined with specific particle formulations to develop robust and cost-effective diagnostic tools. Here we analysed the deposit morphology of drying drops containing polystyrene nanoparticles of different surface properties with human native and mutant hemoglobin. We show that deposit patterns reveal information on both the adsorption of proteins on the particles and their reorganization following adsorption. We then established the conditions that enable to distinguish between native and sickle cell hemoglobin by simple pattern analysis. The suppression of the coffee ring effect and the formation of a disk-shaped pattern is primarily associated to particle neutralization by protein adsorption. However, our findings also suggest that protein reorganization following adsorption can dramatically invert this tendency. Exposure of hydrophobic residues can lead to disk deposit morphologies independently of the global particle charge due to accumulation of particles at the liquid/gas interface during evaporation. This general behaviour opens the possibility to probe protein adsorption and reorganization on particles by the analysis of the deposit patterns, the formation of a disk being the robust signature of particles rendered hydrophobic by protein adsorption. This method is sensitive enough to detect a single point mutation in a protein, as demonstrated by the distinct patterns formed by human native hemoglobin HbA and its mutant form HbS responsible for sickle cell anemia (1). The coffee-ring effect can translate various types of molecular interactions between nanoparticles and proteins into easily distinguishable macroscopic patterns, thus providing valuable insights for the development of future cost-effective diagnostic tools.

Reference

(1) Devineau S, Anyfantakis M, Marichal L, Kiger L, Morel M, Rudiuk S, Baigl D. Protein adsorption and reorganization on nanoparticles probed by the coffee-ring effect: application to single point mutation detection. JACS (2016) 138, 11623-11632



Sickle cell hemoglobin detection in drying drops.jpg

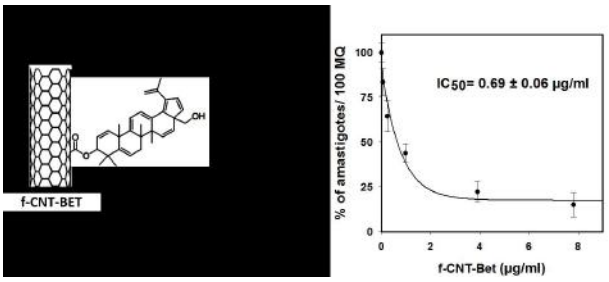
Molecular mechanism and increased antileishmanial activity of carbon nanotube based betulin formulation

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 473

Dr. Prakash Saudagar [1]

1. *National Institute of Technology Warangal*

The present study describes a novel antileishmanial drug formulation of betulin (BET) attached to functionalized carbon nanotubes (f-CNTs) as macrophage targeted drug delivery system. We conjugated BET; a pentacyclic triterpenoid secondary metabolite to carboxylic acid chains on f-CNTs to obtain BET attached functionalized carbon nanotubes (f-CNT-Bet). The fourier transform infrared (FTIR) spectroscopy and transmission electron microscopy (TEM) demonstrated the successful construction of f-CNT-BET. The Bet loading efficiency onto the f-CNTs and Bet release profile were monitored spectroscopically. The drug release profile demonstrated a fairly slow release of Bet. The in-vitro cytotoxicity of BET, f-CNT and f-CNT-BET on J774A.1 macrophage cell line were 211.05 ± 7.14 µg/ml; 24.67 ± 3.11 µg/ml and 72.63 ± 6.14 µg/ml respectively as measured by the cytotoxic concentration required to inhibit 50% cell viability (IC50). The IC50 of BET and f-CNT-BET against intracellular L. donovani amastigote in vitro were 8.33 ± 0.41 µg/ml and 0.69 ± 0.08 µg/ml respectively. The results clearly demonstrating the greater antileishmanial efficiency of f-CNT-BET formulation than BET alone and with no significant cytotoxicity observed on host cells, f-CNT-BET boost new hope for significant application of carbon nanotubes in formulating better drug for leishmaniasis with minimum side effects and high treatment efficacy.



Antileishmanial activity of f-cnt-bet.jpg

At focus compressed 7 and 70 femtosecond pulses for deep tissue multiphoton microscopy

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 489

Dr. Christian Maibohm [1], ***Dr. Francisco Silva*** [2], ***Dr. Edite Figueiras*** [1], ***Dr. Paulo T. Guerreiro*** [2], ***Dr. Marina Brito*** [1], ***Dr. Rosa Romero*** [2], ***Dr. Helder Crespo*** [3], ***Dr. Jana B. Nieder*** [1]

1. *INL-International Iberian Nanotechnology Laboratory,* ***2.*** *Sphere Ultrafast Photonics,* ***3.*** *University of Porto*

We compare the contrast, signal intensities and excitation selectivity of a standard Ti:Sa Tsunami oscillator laser typically used in multi-photon microscopy with about 150 fs pulse duration and compare the performance with an ultrashort laser source with 7 fs short pulses. Pulse compression and measurement was performed at the focus of the objective with a new microscope compatible with the d-scan laser metrology based transmission head.

Using such pulse compression technique we were able to reduce the Tsunami pulse length at the focus of the objective that mainly is affected by the dispersion of the high NA (1.4) microscope objective itself and obtained nominal Fourier Transform limited pulses with 70 fs pulse duration at the position of the sample.

We use a multicolor-labeled 2D FluoCells® (Thermofisher) test sample as well as a pig artery tissue sample to compare contrast, intensity and in addition the deep tissue imaging performance and can report substantial improvements of these parameters using at-focus compressed femtosecond laser pulses. These improvements could play a major role for rendering multiphoton microscopy an excellent tool for noninvasive optical investigation of bioengineered tissues and to drug delivery studies inside tissues.

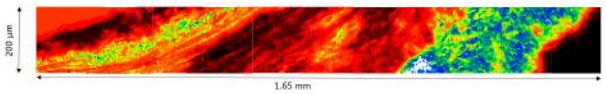


Figure: *Multiphoton image using an ultrashort femtosecond laser source with 7fs pulse duration for a horizontal cut of a pig artery*

Absract picture jpg 2.jpg

In vivo anti-inflammatory activity by trans-resveratrol loaded-solid lipid nanoparticle for skin disorders.

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 553

Ms. Roberta Rigon [1], ***Ms. Maíra Gonçalez*** [2], ***Ms. Camila Rodero*** [2], ***Dr. Marlus Chorilli*** [2]

1. *1Universidade Estadual Paulista "Júlio de Mesquita Filho", Araraquara, Brazil,* ***2.*** *Universidade Estadual Paulista "Júlio de Mesquita Filho", Araraquara, Brazil*

Trans-resveratrol (RES) presents important action to prevent and treat skin disorders. Cutaneous administration of RES has being explored in order to find it on their site of action [1]. Solid lipid nanoparticles (SLN) due to interaction characteristics with skin have being used to drug delivery system for topical application [2]. The aim was verify in vivo anti-inflammatory activity of SLN with RES for cutaneous application therapy. The formulations developed were composed by stearic acid (SA) as solid lipid, polysorbate 80 (P80) and soy phosphatidylcholine (SP) as surfactants, and poloxamer 407 (P407) and glycerin as stabilizers (F1). Cetrimonium bromide (CB) was added as cationic surfactant to promote positive superficial charge (F2). The formulations was add of 0.25% RES (F1.RES and F2.RES). The anti-inflammatory and antinociceptive activities of SLNs were measured by carrageenan-induced paw edema and paw pressure test in mice, respectively [3,4]. This research was approved by Ethics Research Committee of UNESP (n° 68/2015). The average hydrodynamic diameters were 195.0 ± 3.34 nm, 241.3 ± 48.33 nm, 159.15 ± 4.78 nm e 158.25 ± 33.92 nm to F1, F1.RES, F2 e F2.RES, respectively. Zeta potentials (mV) were -25.5 ± 1.01; -26.0 ± 1.67; 30.6 ± 1.13 e 30.0 ± 1.85 mV for F1, F1.RES, F2 e F2.RES, respectively. Entrapment efficacy was analyzed using validated analytical methodology and both formulations (F1.RES and F2.RES) present 50% of RES entrapped. RES solution (1:1 ethanol and water) and F2.RES presented reduction of nociception similar to dexametasone commercial cream, which suggest potent anti-inflammatory activity of RES and F2.RES. Stratum corneum have a negative surface charge that permits greater bioadhesion of positive particles surface, which could explain the increase of antinociceptive activity by F2.RES compared to F1.RES. The results demonstrated the importance to investigate the RES action when it is entrapped in drug delivery system for topical application.

References

[1] Ndiaye, Mary, et al. Archives of biochemistry and biophysics 508.2 (2011): 164-170.

[2] Wissing, Sylvia A., and Rainer H. Müller. International Journal of Pharmaceutics 254.1 (2003): 65-68.

[3] RANDALL, L. O. Arch Int Pharmacodyn, 111 (1957): 409-419.

[4] SANTOS-NOGUEIRA, E. et al. Journal of neurotrauma 29.5 (2012):898-904.

Porous metal–organic-framework MIL-100(Fe) as a nanoscale platform for sustainable release of tetracycline

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 508

Mr. Seyed Dariush Taherzade [1], ***Dr. Aliakbar Tarlani*** [2], ***Dr. Janet Soleimannejad*** [1]

1. *university of tehran,* ***2.*** *Chemistry & Chemical Engineering Research Center of Iran*

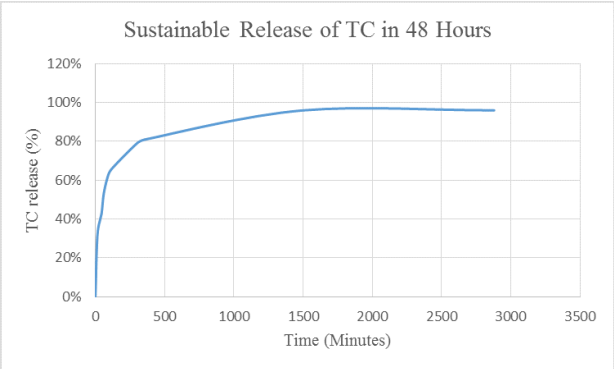
Tetracyclines are a group of broad-spectrum antibiotics used in the treatment of infections of urinary and respiratory tract [1]. Nanoscale porous MIL-100 is a great platform for delivery of broad types of drugs like doxorubicin, ibuprofen and etc [2, 3]. In current research nano MIL-100 was synthesized via Horcajada, et al. method [2]. Sample characterizations carried out with FT-IR, XRD, BET and SEM. Tetracycline (TC) was loaded on MIL-100 in the ratio of 1:3 and the nano-drug-carrier abbreviated as TC@Mil-100. FT-IR spectra confirmed the loading of the drug into the carrier framework. XRD pattern of TC@MIL-100 indicated no pattern of TC. On the other hand, BET analysis showed a 66 percent decrease in available pores and the free surface decreased to 801 Cm3g-1, so it can be concluded that the drug is well dispersed into the nano-pores of MIL-100. The release of TC from MOF was investigated in Simulated Gastric Fluid (SGF) media during 48 hours via UV analysis and it was observed that 96 percent of the drug was sustain released which is an unprecedented amount in comparison with other methods[4]. The results demonstrated the substantially enhanced release of tetracycline as a very slightly soluble drug.

1.Chopra, I. and M. Roberts, Tetracycline antibiotics: mode of action, applications, molecular biology, and epidemiology of bacterial resistance. Microbiology and molecular biology reviews, 2001. 65(2): p. 232-260.

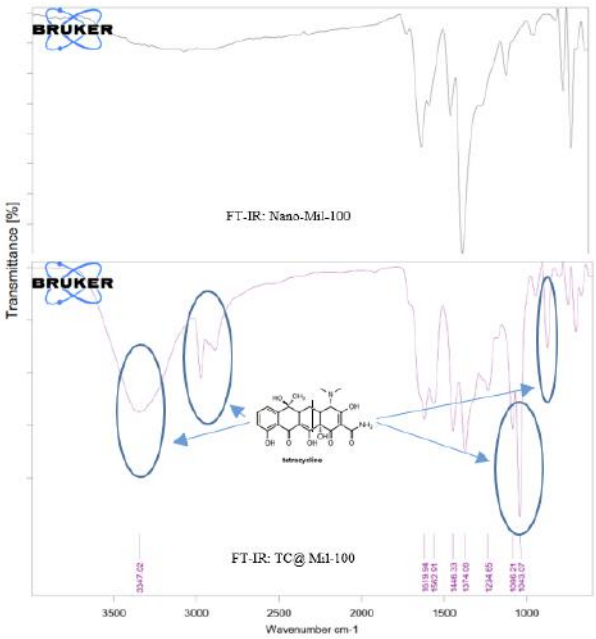
2.Horcajada, P., et al., Porous metal-organic-framework nanoscale carriers as a potential platform for drug delivery and imaging. Nature materials, 2010. 9(2): p. 172-178.

3.Agostoni, V., et al., Impact of phosphorylation on the encapsulation of nucleoside analogues within porous iron (III) metal–organic framework MIL-100 (Fe) nanoparticles. Journal of Materials Chemistry B, 2013. 1(34): p. 4231-4242.

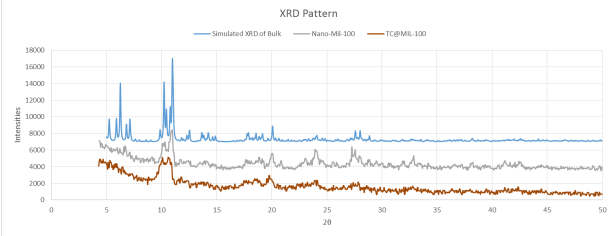
4.Xie, J.F., et al. In Vitro Release Characterization of Tetracycline Hydrochloride from PLGA/Tetracycline Hydrochloride Electrospun Nanofiber Mats. in Advanced Materials Research. 2013. Trans Tech Publ.



Sustainable release of tc in 48 hours.png



Ft-ir comparison.png



Xrd comparison.png

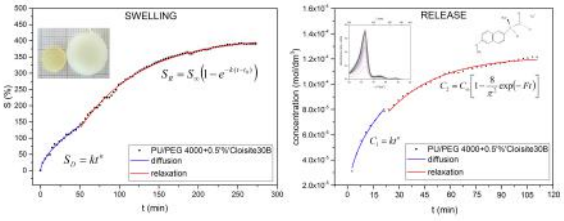
Characterisation of polyurethane nanocomposite hydrogel systems for wound dressing application.

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 552

Ms. Marta Miotke [1], ***Dr. Justyna Strankowska*** [1], ***Prof. Marek Józefowicz*** [1], ***Dr. Michał Strankowski*** [2], ***Prof. Jerzy Kwela*** [1]

1. *University of Gdańsk,* ***2.*** *Gdańsk University of Technology*

In recent years significant efforts have been made by many research groups for designing a new type of wound dressing. Large number of studies concern application of nontoxic, biodegradable polyurethanes which, moreover, are easy accessible and have wide possibilities to change their properties with modification. The goal which we would like to achieve is to obtain wound dressing with capabilities to faster wound healing and alleviation of pain by release of drug by delivery diffusion process. In order to obtain these properties, the process of increase of swelling ability of polymer matrix is important to achieved, for example, by adding nanoparticles which extend the intermolecular spaces in polyurethane matrix. The main purpose of our research is to achieved material with predetermined and well defined hardness, elasticity, and with appropriate swelling and release profiles. In previous papers we described method of synthesis and studies of basic mechanical properties (using DMA technique) and we examined structure of polyurethane nanocomposite by means of X-ray diffraction. The naproxen sodium – commonly used inflammatory drug, was chosen as an active substance potentially released from wound dressing. Before molecule reaches the target receptor, it changes the environment from polar protic – body fluids, to nonpolar cavity receptor. Understanding possible interaction between drug and solvent molecules in the ground and/or excited states was one of the main aim of this work. To achieve this goal, steady-state absorption and fluorescence spectra of naproxen in solvents of different polarities were registered. In addition, using steadystate spectroscopy methods, positive influence of presence of nanoparticles (organofilized montmorillonite – Cloisite®30B) in polymer matrix was observed. Analysis of release active substance from polyurethane matrix were determined basing on light absorption spectra. The concentration of naproxen was registered as a function of time. The most common mechanism of drug release from hydrogels is passive diffusion. In our case this process is guaranteed by presence of nanoparticles. Obtained results confirmed the high possibilities of polyurethane nanocomposite hydrogels as a new type of wound dressing material with the ability to release of drugs.



Graphical abstract.png

Curcumin-loaded cationic solid lipid nanoparticles as a potential platform for the treatment of melanoma

Wednesday, 28th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 550

Ms. Maíra Gonçalez [1], ***Ms. Roberta Rigon*** [1], ***Dr. Marlus Chorilli*** [1]

1. *Universidade Estadual Paulista "Júlio de Mesquita Filho", Araraquara, Brazil*

INTRODUCTION

Curcumin (CUM) presents antineoplastic properties and its incorporation into cationic solid lipid nanoparticles (CSLN) is convenient in order to compartmentalize and delivery this drug to melanoma cells, can be used in melanoma prevention and treatment (1).

METHODS

Formulations preparation

All formulations were produced with Poloxamer 407 (3.2%), cetrimonium bromide (0.7%), water (91.1%), and solid lipids (5%), which were varied obtained F1 composed by stearic acid, F3 composed by Compritol 888 ATO and F2 composed by a blend of both lipids. The ingredients were mixtures and sonicated for 20 minutes at 12-17 W. It were performed determinations of mean hydrodynamic diameter (Dnm), polydispersity index (PdI) and zeta potential (ZP) of each formulation.

Cell viability assay of HaCaT and B16-F10 cells by MTT technique

Healthy keratinocytes cells (HaCaT) and tumorigenic melanocytic cells (B16-F10) were treated with formulations, CUM-free, Doxorubicin solution (PC) and culture medium (NC). After the treatments, the cell viability was evaluated using the MTT technique.

RESULTS AND DISCUSSION

There was obtained systems with Dnm of 218-238 nm, PdI of 0,185–0,350 and ZP of 25,6–30,1 mV.

Viability cells tests showed that the systems developed are more toxic to B16-F10 than HaCat and the incorporation of CUM in CSLNs increase the toxicity of this drug to melanoma cells (B16F10), because the positive charge presented by these systems may promote drug targeting to melanoma cells, that have a high density of negative charge due to high exposure of phosphatidylserine on the surface of cell (2), showing that the CUM-loaded-CSLN are promising for the treatment of melanoma (Figure 1).

(Attach figure 1)

Figure 1. Cell viability assay of HaCaT (a) and B16F10 (b), after exposed to treatments.

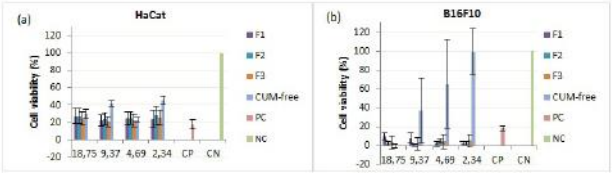
REFERENCES

1. Sharma R.A. et al. Eur J Cancer, 41, 1955–1968, 2005.

2. Sakamoto, J. H. et al., Pharmacol. Res., 62, 57–89, 2010.

ACKNOWLEDGEMENTS

This work was financially supported by Fundação de Amparo à Pesquisa do Estado de São Paulo (FAPESP).



Cell viabity assay jpeg.jpg

Peptide functionalised nanoparticles for the selective induction of apoptosis in target cells

Wednesday, 28th September - 16:00 - Targeted drug delivery and Nanocarriers - Nanomedecine for cancer diagnosis & therapy - Amphitheatre 25 - Oral presentation - Abstract ID: 434

Prof. Mervin Meyer [1], ***Dr. Nicole Sibuyi*** [1], ***Dr. Ntevheleni Thovhogi*** [1], ***Prof. Martin Onani*** [1], ***Dr. Amanda Skepu*** [2], ***Dr. Abram Madiehe*** [3]

1. *University of the Western Cape,* ***2.*** *Mintek,* ***3.*** *Council for Scientific and Industrial Research*

Anti-angiogenic therapies are practical strategies for the treatment of both obesity and cancer. The aim of this study was to develop gold nanoparticles (AuNPs) for targeted anti-angiogenic therapy. The objective was to develop AuNPs that can selectively deliver pro-apoptotic peptides to the endothelial cells of the vascular networks that supply the white adipose tissue (WAT), causing cell death and consequent reduction in the vasculature. Adipose Homing Peptide (AHP) binds to the prohibitin (PHB) which is over expressed on the surface of the endothelial cells in the WAT vasculature of obese individuals. We previously demonstrated that AuNPs functionalized with AHP accumulated in the WAT of animal models of obesity. We evaluated the selective targeting and toxicity of AuNPs that were bi-functionalised with AHP and a pro-apoptotic peptide. We used 3 human cancer cell lines (Caco-2, MCF7 and HT29), of which only one (Caco-2) express PHB. Toxicity was evaluated using the WST-1 and the APOPercentage assays, while the uptake of the nanoparticles was confirmed by ICP-OES and TEM analysis. The cytotoxic activity of the receptor targeted AuNPs was more pronounced in the cells expressing the receptor for AHP. The AuNPs induced significant levels of apoptosis Caco-2 cells, but not MCF7 and HT29 cells. This study shows receptor mediated targeting, and the selective destruction of cells expressing the receptor through the induction of apoptosis and the potential for the development of targeted anti-angiogenic strategy.

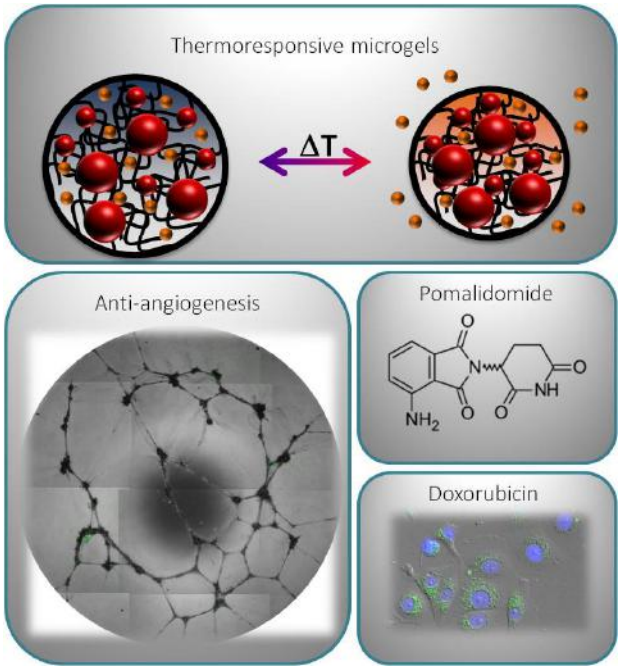
Thermosensitive nanogels with multiple anti-tumour associated effects

Wednesday, 28th September - 16:25 - Targeted drug delivery and Nanocarriers - Nanomedecine for cancer diagnosis & therapy - Amphitheatre 25 - Oral presentation - Abstract ID: 89

Dr. Malou Henriksen-Lacey [1], ***Mr. Malte Strozyk*** [2], ***Dr. Susana Carregal*** [1], ***Prof. Mathias Brust*** [2], ***Prof. Luis Liz-Marzán*** [1]

1. *CIC biomaGUNE,* ***2.*** *University of Liverpool*

Most chemotherapeutic treatments involve combination therapy to inhibit tumour growth, yet the systemic method of administration involving separate drug formulations results in undesirable side effects with a complicated dosing strategy. Consequently, the ability to combine multiple drugs in a single nanoparticulate delivery system in which release is stimulated by external stimuli, thereby diminishing systemic effects, is of interest.
With this in mind we have explored thermosensitive microgels for their ability to change in diameter and volume in response to a change in temperature between 30 – 50°C. Within these vehicles we include gold nanoparticles, which can be used for Raman sensing or local heating, and two drugs of interest for tumour treatment, doxorubicin and pomalidomide. The effects of doxorubicin are well documented whereas pomalidomide remains a relatively novel drug that has shown promising results in the treatment of multiple myeloma (MM), among other actions including the inhibition of angiogenesis and anti-inflammatory cytokines secretion.
Gold nanoparticles, doxorubicin and pomalidomide were all entrapped within the microgels, after which positive or negatively charged polyelectrolytes were used as a coating material to stabilise the particles. In vitro experiments using single, double or unloaded microgels were conducted to assess the cytotoxic and anti-angiogenic effects of doxorubicin and pomalidomide respectively. The EPR effect of cancer cells was investigated using an in vitro mixed cell co-culture model.
Collapsed states of the microgels were observed at biological relevant temperatures which led to release of both doxorubicin and pomalidomide. Encapsulation did not affect the activity of the drugs. In vitro studies with a variety of cell lines showed high levels of microgel uptake and a slower release compared to free drug. Pomalidomide was shown to have various effects including the inhibition of LPS-induced cytokine production, disregulation of the actin cytoskeleton and inhibition of angiogenesis in tube-formation assays.
We show that these thermosensitive microgels are highly flexible tools for the encapsulation of a variety of drug or sensing compounds which importantly remain active upon release. Due to the inclusion of gold nanoparticles the microgels also can be used as sensing particles or be stimulated with light to induce local heating.



Mhenriksen.iconan2016.jpg

Promising New NanoTheranostic Quantum dots Based on Ag2S-PEG-FA

Wednesday, 28th September - 16:50 - Targeted drug delivery and Nanocarriers - Nanomedecine for cancer diagnosis & therapy - Amphitheatre 25 - Oral presentation - Abstract ID: 568

Prof. havva yagci acar [1]

1. *Koc University*

Introduction

There is a tremendous effort in the development of nontoxic QDs emitting in the near-infrared (NIR) for medical use, both as an optical imaging and as gene/drug delivery agents. This provides an opportunity for deep tissue penetration and reduction of the tissue autofluoresence. Other important issues in practical application of QDs are the overall size, blood circulation, molecular targeting and stability. Ag2S quantum dots emerged as the most promising QDs in the last 5 years. They emit in the NIR region and are highly cyto-hemocompatible. Here, we report the first direct and size tuned synthesis of PEG coated Ag2S NIR QDs and their theranostic use for optical imaging and folic acid (FA) targeted DOX delivery to HeLa cells.

Methods

Ag2S, Na2S and MPEG-SH were dissolved in DIW under Argon at different ratios and at pH values between 3 or 7.5. Reactions were run at 90°C. For FA tagging 30mol% HOOC-PEG-SH was used in the coating and FA was conjugated with EDC/NHS. DOX.HCl was loaded to QDs at pH 6. MTT assay was used to analyze cytotoxicity of particles in HeLa and NIH-3T3 cells.

Results and Discussions

In the size and emission tunning of QDs, increasing Ag/S decreased the particle size. Synthesis at acidic pH produced larger QDs with stronger emission. Increasing PEG molecular weight from 2000 Da to 5000 Da increased the QY about 40 % with 43 nm red shift in peak maxima. All QDs were below 50 nm. Between 5-25 g/mL Ag dose, no significant cytotoxicity was observed in either cell lines. Folate targeted and DOX loaded particles delivered DOX to the cells more effectively than untagged QDs. About 50% drop in the viability with only 15 nM DOX was achieved with FA targeting.

Conclusions

Here, in a simple one step aq. synthesis, PEGylated Ag2S NIR QDs were synthesized with emission maxima between 847-930 nm with QY as high as 65 %, using HS-PEG coating. These particles were non-toxic to HeLa and NIH/3T3 even at mg range. Internalized particles, generates a strong NIR signal. FA-tagged particles showed excellent drug efficiency coupled with receptor mediated uptake.

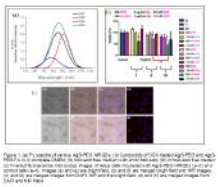


Figure 1.png

Study of RNA interference mediated by lipid-coated calcium phosphate nanoparticle transfection in high-grade gliomas.

Wednesday, 28th September - 17:15 - Targeted drug delivery and Nanocarriers - Nanomedecine for cancer diagnosis & therapy - Amphitheatre 25 - Oral presentation - Abstract ID: 140

Dr. Laura Pandolfi [1], ***Dr. Miriam Colombo*** [1], ***Mrs. Benedetta Santini*** [1], ***Mrs. Lucia Salvioni*** [1], ***Dr. Svetlana Avvakumova*** [1], ***Prof. Silvia Nicolis*** [1], ***Prof. Davide Prosperi*** [1]

1. *University of Milano - Bicocca*

Gliomas are the most prevalent brain tumor types in adults. The recent discoveries in gene regulation involved in gliomas initiation and development allowed for the generation of a new therapeutic approach using a sequence-specific gene silencing to alter gene activities.1,2

Calcium phosphate nanoparticles (CaP NPs) have been shown to be an attractive tool for non-viral gene delivery thanks to their biocompatibility and biodegradability. Indeed, calcium ions form complexes with the nucleic acid backbone, thus stabilizing RNA and DNA structures and, therefore, avoiding their fast degradation by nucleases.3,4

As a model platform useful for the investigation of gene silencing in gliomas, we synthesized CaP NPs for siRNAs delivery in GFP-expressing T794 oligodendroglioma cells isolated from transgenic new born mice induced with high grade brain tumor. This ex vivo model allowed us to study the efficiency of anti-GFP siRNA delivery following GFP fluorescence relating to the relevant gene expression. In order to enhance the circulation half-life in future in vivo experiments and to allow the functionalization of CaP nanoparticles, the core was coated with a cationic asymmetric lipidic bilayer (LCP NPs). LCP NPs were characterized by transmission electron microscopy (14.3±1.8 nm), dynamic light scattering and zeta-potential, showing a mean hydrodynamic diameter of 33.2±3.8 nm and a mean zeta-potential of +31.95±1.3 mV.

Interestingly, both control (empty LCP NPs) and siRNA-loaded LCP NPs did not exert a cytotoxic effect on T794 cells in 0.2-7 nM concentration. Flow cytometry analysis showed huge internalization of LCP NPs (2 nM) by cells up to 24 h of incubation. Treatment with siRNA-loaded LCP NPs confirmed their efficiency by decrease of the mean fluorescence intensity of GFP signal. On the basis of these promising results, we can translate this non-viral gene delivery system to alter other oncogenes supposed to play a pivotal role in gliomas initiation and development, including Sox2.

Nanometronomics: doxorubicin-loaded H-ferritin allows for tailored treatment of breast cancer based on lower doses and higher safety

Wednesday, 28th September - 17:40 - Targeted drug delivery and Nanocarriers - Nanomedecine for cancer diagnosis & therapy - Amphitheatre 25 - Oral presentation - Abstract ID: 106

Dr. Serena Mazzucchelli [1], Ms. Michela Bellini [2], Dr. Marta Truffi [1], Dr. Luisa Fiandra [1], Ms. Maria Antonietta Rizzuto [2], Dr. Luca Sorrentino [1], Dr. Fabio Corsi [1], Prof. Davide Prosperi [2]

1. *Università degli Studi di Milano,* ***2.*** *Università degli Studi di Milano-Bicocca*

The standard clinical approach for cancer chemotherapy is based on the concept of maximum tolerated dose (MTD) of drug for a patient. This strategy, however, presents several disadvantages, including major systemic toxicity and development of therapeutic resistance due to prolonged time between treatments. In addition, solid tumors are less effectively eradicated by MTD-based treatments because their proliferation is also sustained by the host microenvironment. Metronomic chemotherapy is based on more frequent and lower dose drug administrations compared to MTD [1]. Although initially intended to prevent angiogenesis, recent evidence suggests that potential new mechanisms of action are involved, including restoration of anticancer immune response and induction of tumor dormancy. For this reason, metronomic chemotherapy has gradually gained interest among clinicians for either primary systemic or maintenance therapy. However, low drug accumulation at the tumor and poor effectiveness against highly aggressive metastatic cancer limit the clinical application. To improve the efficacy of metronomic chemotherapy, we have investigated the potential of recombinant H-ferritin (HFn) nanocages targeted delivery of lower doses of chemotherapeutic drugs, such as doxorubicin (DOX). HFn was produced by genetic engineering and loaded with DOX [2]. Mice bearing a highly aggressive, DOX-resistant 4T1 breast cancer cells were treated in parallel with placebo, free DOX and HFn-DOX and monitored until day 21 at repeated doses as low as 1.24 mg/Kg, significantly lower than the minimal clinical dosage (2.4 mg/kg). Our results highlight that metronomic treatment with nanoformulated DOX is able to inhibit cancer growth with a lower dose and a significant improvement in DOX toxicity profile, even in a highly metastatic DOX-resistant breast cancer model in vivo. Histological analysis of cardiac tissues suggested that HFn-DOX allows overcoming cardiotoxicity, one of the most severe side effects of DOX. In conclusion, HFn-DOX has a tremendous potential for the development of novel nanometronomic chemotherapy for the next generation of safe and tailored oncological treatments.

Turn-on and Color-changeable Fluorogenic Sensor Created by the 10BASEd-T

Wednesday, 28th September - 16:00 - Biological & medical nanodevices and biosensors - Tower 24 - Room 101 - Oral presentation - Abstract ID: 151

Prof. Masumi Taki [1]

1. *The University of Electro-Communications (UEC)*

6-propionyl-2-(dimethylamino)naphthalene (PRODAN) is a well-known solvatochromic fluorescence reporter which shows both turn-on and color-changing effect when exposed in a hydrophobic environment [1]. Because of its extreme small size with lipophilic/nonionic property, we hypothesize that it can be interacted weakly with various hydrophobic pockets of most target biomacromolecules such as proteins. A conjugation of an appropriate surrounding structure to PRODAN core would strengthen both specificity and affinity to the target along with the solvatochromism, and consequently create a target-specific fluorogenic sensor.

Meanwhile, we have established a library construction system, namely gp10 based-thioetherification (10BASEd-T), by conjugation between artificial molecules as the cores and randomized library peptides as the surroundings [2, 3]. By using this system, PRODAN core was conjugated with randomized peptide library on T7 phage, and selection against a target protein (glutathione-S-transferase; GST) was performed. After several rounds of biopanning, a consensus sequence of NTVSC*HGF (C* represents PRODAN-hybridized Cys) was found. The chemically synthesized hybrid showed intense blue-cyan fluorescence only when GST was present. After the addition of excess amounts glutathione (GSH; a natural ligand of GST), it disappeared and weak yellow fluorescence appeared instead. Detailed results such as obtaining dissociation constant, thermodynamic parameters, and epitope-mapping upon the specific interaction between the hybrid sensor and GST will also be presented [4].

[1] Weber, G., Farris, FJ. (1979) Biochemistry, 18, 776–9.

[2] Fukunaga, K., Hatanaka, T., Ito, Y., Taki, M. (2013) Mol. BioSyst., 9, 2988–91.

[3] Fukunaga, K., Hatanaka, T., Ito, Y., Minami, M, Taki, M. (2014) Chem. Commun., 50, 3921–3; inside cover article.

[4] Taki, M., Inoue, H., Mochizuki, K., Yang, J, Ito, Y. (2016) Anal. Chem., 88, 1096–99 (2016).

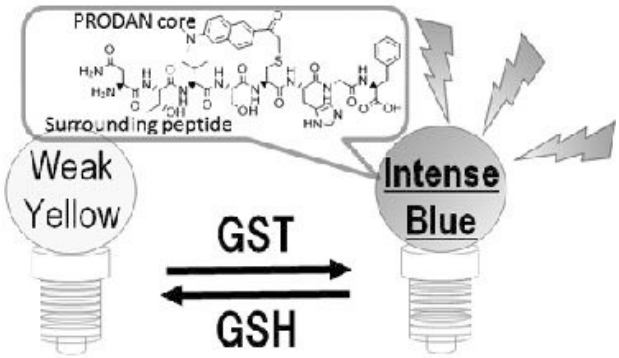

PRODAN core
Surrounding peptide
Weak
Yellow
GST
GSH
Intense
Blue

Image.jpg

One-step DNA Detection through Dual-Color Confocal Analysis of DNA-Assembled Scattering Nanoparticles: Case of a Fragment of Sesame

Wednesday, 28th September - 16:25 - Biological & medical nanodevices and biosensors - Tower 24 - Room 101 - Oral presentation - Abstract ID: 265

Dr. Stephanie VIAL [1], ***Mr. Youri BERRAHAL*** [1], ***Dr. Marta PRADO*** [2], ***Dr. Jérome WENGER*** [1]

1. *Institut Fresnel, Aix Marseille Université,* ***2.*** *International Iberian Nanotechnology Laboratory*

DNA-based detection methods have many applications in clinical diagnosis, food science, and environmental control. However, most of the analytical techniques currently in use, such as PCR, are expensive, cumbersome, and time consuming. Thus, methods such as enhanced surface plasmon resonance and fluorescence-based devices have been developed, but still present some limitations. Here, we present a novel approach based on Photon Cross Correlation Spectroscopy (PCCS) of metallic nanoparticles (NPs) that provides increased sensitivity, rapid detection and reduced cost in order to open new and breakthrough features for diagnosis.

The detection of DNA relies on the self-assembly of two distinct scattering NPs (silver nanoparticles (AgNPs) and gold nanorods (AuNRs)) mediated via specific-based pair recognition between ss-DNA target and ss-DNA probes anchored to NPs. Upon illumination using two laser beams, the particles passing through the volume of analysis are excited and their resulting scattered lights are collected and analyzed together. In the absence of target, both NPs move independently and their signals are not correlated. However, as the presence of DNA target initiates aggregation of the NPs through hybridization, AgNPs and AuNRs thus diffuse simultaneously. Consequently, their scattering responses are correlated, giving rise to temporal coincidence (Figure 1).

To validate the developed proof-of-concept PCCS, the study focused on the detection of a specific fragment of sesame, an allergenic food ingredient. Here, we have considered several aspects with the aim to improve detection efficiency within the shortest time. The optimization of the hybridization events between a target DNA and its probes, the sensitivity with quantitative analysis and the specificity to particularly distinguish perfectly matched DNA from defective one have been investigated. This new detection method was able to in-situ detect and quantify specific DNA sequence of sesame in a range of concentration from 5 pM to 1.5 nM , with LOD of 1 pM, after 30 min of incubation at 65°C. In addition, we have observed a net decrease in the detection when using one single matched and one deleted base in the sequence of the target and no possible detection in presence of non-complementary DNA

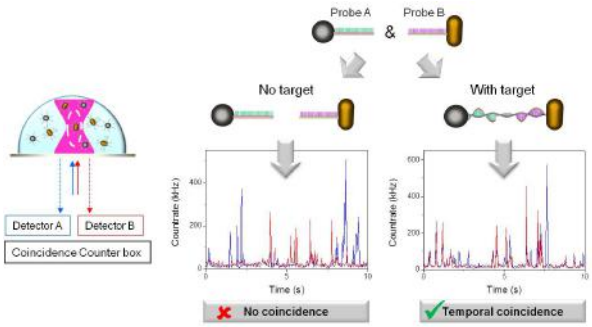

Probe A
&
Probe B
No target
With target
Detector A
Detector B
Coincidence Counter box
Countrate (kHz)
Time (s)
No coincidence
Temporal coincidence

Image2.jpg

Up-converting and down-converting nanoparticle-based aptasensor model for multiplex detection of foodborne pathogens

Wednesday, 28th September - 16:50 - Biological & medical nanodevices and biosensors - Tower 24 - Room 101 - Oral presentation - Abstract ID: 375

Dr. Hasan Kurt [1], ***Dr. Meral Yüce*** [1], ***Mr. Babar Hussain*** [1], ***Prof. Hikmet Budak*** [1]

1. *Sabanci University*

Advances in the selection of synthetic affinity agents, called aptamers, have provided us with the opportunity to develop highly efficient and specific biosensors. Coupling of aptamers with fluorescent nanoparticles have led to the rapid detection of various molecules from ions to disease biomarkers (1). However, only one type of nanoparticle has been used in these multiplexed assays. Herein, we report a multiplex sensing method based on the unique excitation profiles of two down-converting quantum dots (QD) and two up-converting nanoparticles (UCNP) that are coupled with the target-specific DNA aptamers. Our group recently reported the dual-excitation strategy for detection of Salmonella typhimurium and Staphylococcus aureus (2). In the current work, the dual excitation method is extended to detection of four foodborne pathogens, S. typhimurium, S. aureus, Listeria monocytogenes, and Pseudomonas aeruginosa.

Pathogen-specific aptamers were coupled with CdTe QDs at 510 and 720 nm emissions, and NaYF4-based UCNPs at 545 and 800 nm emissions. The resulting nanoparticles were conjugated with the magnetic beads that were previously coupled with the corresponding partial complementary DNA sequences. These conjugates were simultaneously used as molecular recognition elements for the detection. Partially complementary ssDNA-decorated magnetic beads were employed in the sensing system for separation purposes. Aptamer coupled QD and UCNP conjugates, and cDNA decorated magnetic beads were characterized using UV-Vis spectroscopy, Circular Dichroism, and Dynamic Light Scattering techniques. The multiplex detection of the selected food-borne pathogens was implemented using consecutive 335 nm and 980 nm excitations that eliminated the signal overlap between the nanoparticle species. Dual-excitation multiplex aptasensor enabled detection of the tar-gets at trace amounts. The absence of signal cross-talk between the nanoparticles rendered multiplex detection of foodborne pathogens, offering a promising alternative to the conventional methods for detection.

Acknowledgment This project is supported by The Scientific and Technological Research Council of Turkey, TUBITAK Grant ID: 114Z379 (Cost Action CM1403).

1.Yüce, M., Ullah, N. & Budak, H. Trends in aptamer selection methods and applications. Analyst 140, 5379–5399 (2015).

2.Kurt, H., Yüce, M., Hussain, B. & Budak, H. Dual-excitation upconverting nanoparticle and quantum dot aptasensor for multiplexed food pathogen detection. Biosens. Bioelectron. 81, 280–286 (2016).

Analysis of the controlled drug release (CDR) from biopolymer nanoparticles during the initial burst using a novel modeling method

Wednesday, 28th September - 17:15 - Biological & medical nanodevices and biosensors - Tower 24 - Room 101 - Oral presentation - Abstract ID: 141

Ms. Cristiana de Azevedo [1], ***Dr. Moritz Von Stosch*** [2], ***Prof. Rui Oliveira*** [1]

1. *Universidade Nova de Lisboa,* ***2.*** *University of Newcastle*

In the initial stage of the controlled release of a drug from a nanoparticle into a medium a phenomenon referred to as burst can occur. During burst a large amount of drug is released over a small period of time. Apart from the loss in the overall CDR time of actuation, high initial drug release rates can result into toxic drug levels, which would not be attained otherwise. The initial burst has been studied in the past, but with little success in elucidating the mechanisms that control the phenomenon.

In this contribution, a mathematical model is established to investigate how experimental conditions and nanoparticle formulations impact on the initial burst release. Experimental conditions, nanoparticle formulations and drug release profiles were extracted from publications for drug-PLGA or -PLGA/PEG carriers and a database was created. Subsequently, statistical methods were utilized to analyze the data and a model was developed that can predict the burst release based on experimental conditions and nanoparticle formulations. Good agreement between model predictions and experimental burst data was obtained. Further analysis revealed that a clear augmentation in the quantity and kinetics of the burst is obtained when PEG is bound to PLGA. It also seems that an increase in the burst release quantity occurs for greater carrier particles, i.e. going from 5E1 nanometers towards microparticles, but at a slower rate. Longer chains of PLGA and smaller drug molecules show an enhancement on the burst rate.

The increased understanding of the burst release can in future be used to manipulate the system more rationally e.g. to reduce the intensity of the burst release.

Detection of cytokines using biosensor based on functionalized nanocarriers growth on gold electrode

Wednesday, 28th September - 17:40 - Biological & medical nanodevices and biosensors - Tower 24 - Room 101 - Oral presentation - Abstract ID: 96

Prof. Abdelhamid Errachid [1], ***Ms. Sahar Chaibi*** [2], ***Dr. toufik hadjersi*** [3], ***Mr. Abaidia Seddik El Hak*** [2], ***Dr. Abdoullatif Baraket*** [1], ***Prof. Joan Bausells*** [4], ***Dr. yaakoubi nourdin*** [5], ***Dr. Nadia Zine*** [1]

1. *Université Claude Bernard Lyon1,* ***2.*** *Université M'hamed Bouguera,* ***3.*** *Unité de recherché UDTS/CRTSE,* ***4.*** *Instituto de Microelectronica de Barcelona, IMB-CNM (CSIC), Campus UAB, 08193, Bellaterra, Spain.,* ***5.*** *Université du Maine*

Nowadays , it is well established that the performance of biosensors depends greatly on the influence imposed on biomolecules by immobilisation.In this context, the use of nanomaterials for the construction of biosensing devices constitutes one of the most interesting approaches. Application of gold nanoparticules in biosensing has attracted considerable attention because gold nanoparticules (GNPs) are the most compatible nanomaterial for preparation of engineered nanoplatforms in smart sensing devices beside the fact that they have interesting electronic , optical ,thermal and catalytic properties . however, their application is obviously affected by many conditions, such as the size , the shape , the roughness of the substrate and the density of the particules on the surface . In this work we will present some phenomena noticed linked to the impact of gold Np characteristics's on the sensing of cytokine. The cytokine studied for this purpose were : IL1β, IL-10 and IL-6 , using a spectroscopy of impedance for the detection of their corresponding antigens in the range of 1 pg/ml to 30 pg/ml. Effect such as antibodies/Np ratio on the sensing was noticed and this has affected really the sensibility of the biosensor. The gold nanoparticules were electrochemicaly synthesized from KAu(CN)2 diluted into phosphate buffer (PBS) at acidic bath using the chronoamperometric.

The sensitivity of Au-nanoparticles modified biosensor was more pronounced than gold biosensor. The limit of detection (LOD) of the gold biosensor was 1.5 pg/mL. The plot was linear (r=0.9968) within the range of 1-15 pg/ml. Whist the LOD of Au-nanoparticles modified biosensor was 1 pg/mL. The sensitivity of this modified biosensor was enhanced by the introduction of gold nanoparticules as electrolyte/gold interface has been increased. This biosensor is very promising for a rapid and sensitive detection.

Bio-compatibilised carbon nanotubes display significant anti-tumoral effects in solid melanomas

Wednesday, 28th September - 16:00 - Nanomedecine for cancer diagnosis & therapy - Tower 24 - Room 103 - Oral presentation - Abstract ID: 302

Dr. Mónica L. Fanarraga [1], ***Mrs. Eloisa González-lavado*** [1], ***Dr. Lorena García-hevia*** [1], ***Ms. Carmen Pesquera*** [1], ***Mr. Fernando González*** [1], ***Dr. Jesús González*** [1], ***Dr. Juan C Villegas*** [1], ***Dr. Rafael Valiente*** [1]

1. *Universidad de Cantabria-IDIVAL*

Multiwalled Carbon Nanotubes (MWCNTs) penetrate inside cells binding microtubules and interfering with the cellular biomechanics (1,2). This interaction leads to the formation of biosynthetic tubulin polymers (3,4) that display an enhanced stability, and this triggers anti-proliferative (4,5), anti-migratory (6) and cytotoxic (7) effects in different types of cancer cells, leading to antineoplastic effects in solid melanoma tumors (8) in murine models. Moreover, this intrinsic CNT antitumor activity is complementary -and synergetic- to that of traditional microtubule-stabilizing anticancer drugs such as taxol® (paclitaxel)(8). On the other hand, as is also the case for traditional cytotoxic chemotherapy, CNTs can also interfere with the function of heathy cells and produce many unwanted side-effects (9). Consequently, unless most concerns about the toxicity of these materials disappear, the development new treatments based on CNTs offer a poor risk-to-benefit ratio in oncology. Here we demonstrate how mild MWCNTs oxidative treatments enhance intracellular degradation by macrophages while still maintaining the ability of these nanomaterials to interfere with cellular cytoskeletal elements triggering anti-proliferative and cytotoxic processes in cultured cancer cells. Furthermore, we show how single intratumoral dosages of o-MWCNTs produce significant anti-tumoral effects in vivo, in solid malignant melanomas produced by allograft transplantation in murine recipients. We believe these findings have critical implications for the development of new CNT-based nanotherapies to overcome drug resistance in cancer among other applications.

References

1L. García-Hevia,et al. Nanomedicine, 2014, 9, 1581–1588.

2C. Dong, et al. J. Mater. Chem. B, 2015, 3, 3983–3992.

3C. Z. Dinu, et al. Small, 2009, 5, 310–315.

4L. Rodriguez-Fernandez, et al. ACS Nano, 2012, 6, 6614–6625.

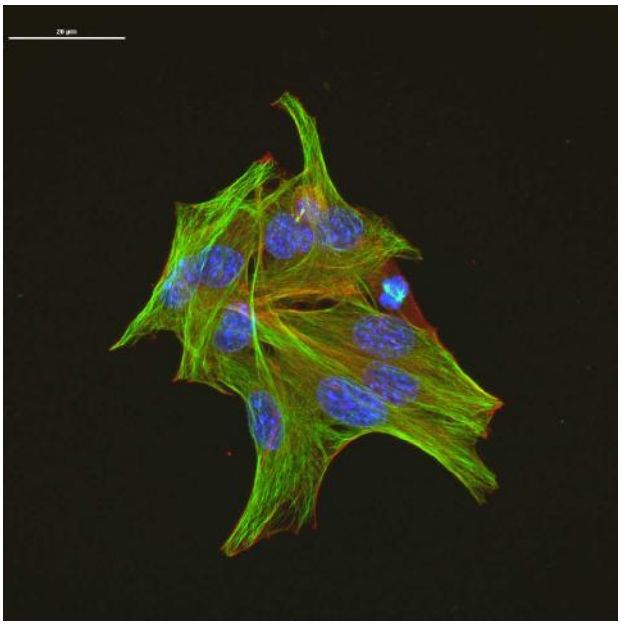
5L. M. Sargent, et al. Mutat. Res. - Genet. Toxicol. Environ. Mutagen., 2012, 745, 28–37.

6L. García-Hevia, et al. Adv. Healthc. Mater., 2015, 4, 1640–1644.

7L. García-Hevia, et al. Curr. Pharm. Des., 2015, 21, 1920–29.

8L. García-Hevia, et al. Adv. Healthc. Mater., 2016, 9:1080-7.

9K. Kostarelos, Nat. Biotechnol., 2008, 26, 774–6.



B16f10 mw24h 20ug 3.jpg

Targeting of TRAIL conjugated maghemite nanoparticles for biomedical applications

Wednesday, 28th September - 16:25 - Nanomedecine for cancer diagnosis & therapy - Tower 24 - Room 103 - Oral presentation - Abstract ID: 404

Mrs. Hanene BELKAHLA [1], ***Dr. Myriana Hemadi*** [2], ***Dr. Guillaume Herlem*** [1], ***Dr. Olivier Micheau*** [3], ***Prof. Souad Ammar*** [2], ***Prof. Tijani Gharbi*** [1]

1. *Nanomedicine Lab, Imagery & Therapeutics, EA 4662, Université de Franche-Comté, Besançon, France.,* ***2.*** *ITODYS, Université Paris Diderot, SPC, CNRS UMR-7086, Paris, France,* ***3.*** *INSERM, UMR866, Lipides nutrition cancer, Facultés de Médecine et de pharmacie, Université de bourgogne, Dijon, France*

Iron oxide nanoparticles (NPs) have become increasingly important for cutting-edge applications in biomedicine due to their biocompatibility and attractive magnetic properties. Super-paramagnetic maghemite nanocrystals possess high magnetic susceptibility, low remanence, low coercivity, and high saturation magnetization. These properties make them ideal candidates for various biomedical applications including magnetic resonance imaging (MRI), magnetically controlled drug delivery (MCDD) and magnetic hyperthermia (MH) [1]. The use of these NPs for in-vivo diagnostics and/or therapy assays requires their coupling through covalent bonding to bioactive molecules (antibodies, enzymes, proteins, DNA, etc.).

TRAIL is a member of the tumor necrosis factor (TNF) superfamily. This protein is able to specifically bind to its receptor which is expressed at the surface of cancer cells and induces cell death [2].

Maghemite nanoparticles (MNP) with diameter of 10 and 100 nm were synthetized by the polyol method, coated with (3-aminopropyl)triethoxysilane (APTES)[3] and coupled to TRAIL protein. The ratio of TRAIL per NP was estimated by Prussian blue and protein assay. The magnetic properties were checked by SQUID before and after grafting. The efficiency of the targeting was checked in cellulo in different cell lines (HCT116, HEPG2, etc.). The resulting magnetic nanovector (NV) seems to be a good candidate for biomedical applications such as cancer therapy.

1.Laurent, S., et al., Magnetic fluid hyperthermia: focus on superparamagnetic iron oxide nanoparticles . Adv Colloid Interface Sci, 2011. 166(1-2): p. 8-23.

2.Zakaria, A.B., et al., Nanovectorization of TRAIL with single wall carbon nanotubes enhances tumor cell killing. Nano Lett, 2015. 15(2): p. 891-5.

3.Piraux, H., et al., Nanoparticles and the Transferrin-Receptor-1 iron-Acquisition Pathway – Synthesis, Kinetics, Thermodynamics and Cellular Internalization of a Holotransferrin-Maghemite Construct. . Biochim. Biophys. Acta. , 2013. 1830 p. 4254–4264.

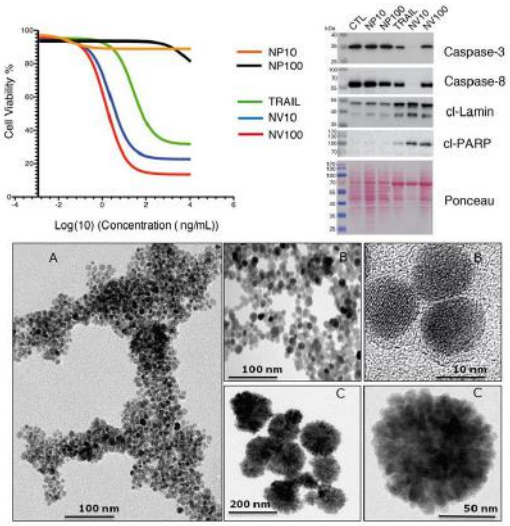


Figure:

- Cell viability of HCT116 cells to TRAIL, non functionalized MNP and TRAIL functionalized MNP (MNP@TRAIL).
- Cell extracts from HCT116 cells stimulated or not with MNP,TRAIL or MNP@TRAIL were processes for immunoblot with antibodies to pro-caspase 3, pro-caspase 8, cleaved Lamin and cleaved PARP. Ponceau staining is shown below, Molecular weight markers are shown on the left handside.
- Transmission Electron Microscopy (TEM) of the produced MNP sized 5, 10, 100 nm .

Iconan 2016.jpg

New promising Glucose-Metal Nanoparticles for potential applications in Radiotherapy

Wednesday, 28th September - 16:50 - Nanomedecine for cancer diagnosis & therapy - Tower 24 - Room 103 - Oral presentation - Abstract ID: 63

Dr. Francesco Porcaro [1], ***Prof. Chiara Battocchio*** [2], ***Prof. Antonio Antoccia*** [1], ***Dr. Ilaria Fratoddi*** [3], ***Dr. Iole Venditti*** [3], ***Dr. Anna Fracassi*** [1], ***Dr. Igor Luisetto*** [1], ***Dr. Andrea Ugolini*** [1], ***Prof. Maria Vittoria Russo*** [3], ***Prof. Giovanni Polzonetti*** [2]

1. *Roma Tre University,* ***2.*** *University of Rome "Roma Tre",* ***3.*** *Sapienza University of Rome*

Recently, a novel approach in radiotherapy consists in the use of “High Z materials” for increasing radiation dose absorption in tumour site and therefore improving the effects of radiotherapy. Metal Nanoparticles (NPs) like Gold (Z=79) are the most promising candidate to become suitable materials for standard treatment in radiotherapy, due to their efficacy in delivering huge amounts of high Z atoms in the proximity of cancer cells nucleus [1]. Despite the great success of nanoparticles in this framework, there is still need to open new routes for the synthesis of more stable and functional nanoparticles.

The synthesis, characterization and assessment of biological response of innovative negatively charged functionalized metal nanoparticles is herein reported, for potential applications in the field of radiotherapy and drug delivery. Gold, Platinum and Silver nanoparticles (AuNPs, AgNPs, PtNPs, respectively) functionalized with two capping agents: the 3-mercapto-1-propansulfonate (3-MPS) and 1-β-thio-D-glucose (TG), have been on purpose synthesized and fully characterized. Advanced characterization techniques including X-Ray Photoelectron Spectroscopy (XPS) were applied to probe the chemical structure of the synthesized nanomaterials. Z-potential and Dynamic Light Scattering measurements allowed assessing the nanodimension, dispersity, surface charge and stability of Nanoparticles. , in order to investigate the nanoparticles-cells interaction and evaluate the uptake efficiency for AuNPs, Transmission Electron Microscopy (TEM) and Flame Atomic Absorption Spectroscopy (FAAS) were performed to the "in vitro" Human Salivary Gland (HSG) cell model. Moreover, a deep cytotoxicity evaluation was assessed by means of MTT and SRB assays [2]. Preliminary data on NPs radiosensibilization were obtained by means of clonogenenic assay. In conclusion, with the purpose of increasing the amount of metal atoms inside the cell we have optimized the synthesis for a new kind of biocompatible and stable negatively charged TG-functionalized nanoparticles. Such particles are very promising for radiotherapy application.

[1] Neshatian M., Yang C., Hegarty N., Chithrani B. D., Precious metals for Biomedical Applications, Chapter: 4, Woodhead Publishing Baltzer N., pp.87-106 2014

[2] Porcaro F., Battocchio C., Antoccia A., Fratoddi I., Venditti I., Fracassi A., Luisetto I., Russo MV., Polzonetti G. Colloids Surf B Biointerfaces. 2016

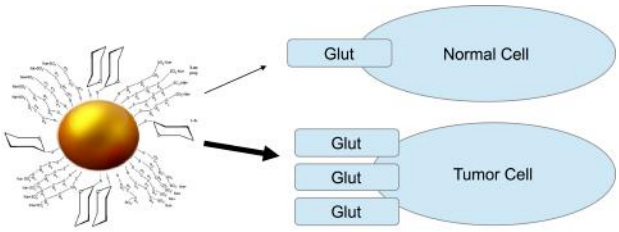


Figure Example of Gold Nanoparticle functionalized with TG and 3MPS. Glucose nanoparticles are internalized most by tumour cell due to glucose receptor overexpression.

Gnp.jpg

Microfluidic synthesis and biological evaluation of photothermal biodegradable copper sulphide nanoparticles

Wednesday, 28th September - 17:15 - Nanomedecine for cancer diagnosis & therapy - Tower 24 - Room 103 - Oral presentation - Abstract ID: 269

Ms. Isabel Ortiz de Solorzano [1], ***Mr. Martín Prieto*** [1], ***Dr. Gracia Mendoza*** [1], ***Dr. Teresa Alejo*** [1], ***Dr. Silvia Irusta*** [1], ***Dr. Víctor Sebastián*** [1], ***Dr. Manuel Arruebo*** [1]

1. *University of Zaragoza*

INTRODUCTION

Copper(II) sulfide(CuS) nanoparticles are semiconductor chalcogenides with electronic and optical properties. As plasmonic material, it absorbs near infrared light converting it into heat due to the excitation of direct and indirect transitions and plasmonic photoexcitation(1)(Smith, 2015). Those optical properties make them useful in a wide variety of biomedical applications.

Biodegradability and higher photothermal conversion efficiencies(1) are one of the main advantages of CuS nanoparticles compared to other plasmonic nanoparticles. Thus, Guo et al.,2013, demonstrated that plasmonic hollow gold NPs remained in the body one month after injection in BALB/c mice at high levels(> 96%)whereas, only a 10% of CuS NPs injected remained in the animals, being mostly excreted following the hepatobiliary route.

Polydispersity, low-yield and batch-to-batch inconsistencies are the main shortcomings when synthesizing NPs which can be overcome by using microfluidic reactors. In this work we have produced CuS NPs in a continuous process showing high photothermal efficiency as well as with the ability of generating reactive oxygen species(ROS).

METHODS

CuS NPs were synthetized using a batch reactor at different temperatures following the work of Ramadan et al.2012. The continuous microfluidic synthesis was carried out by using two consecutive Y-shaped PEEK micromixers to produce sacrificial Cu2O NPs in a first step and CuS NPs in a second step. The NPs obtained were characterized by TEM, XRD, UV-vis, XPS and DLS. Photothermal effects and ROS generation were also evaluated. The NPs cytotoxicity and evaluation of cell apoptosis were performed on mouse mesenchymal stem cells, human dermal fibroblasts and THP1 human monocytes and macrophages.

RESULTS AND DISCUSSION

It is possible to synthesize aqueous CuS NPs in a continuous manner showing absorbance in the near infrared region of the electromagnetic spectrum with a microfluidic reactor. The NPS physicochemical properties are similar to the ones obtained in the conventional batch synthesis although the synthesis times were reduced 4 fold when using the microreactor. Under simulated physiological conditions CuS nanoparticles degrade into soluble copper sulfates which highlights the advantage of those nanomaterials compared to other conventional plasmonic nanomaterials. Those NPs show at subcytotoxic doses an elevated photothermal effect as well as ROS generation.

Development of Innovative Multistage Nanovectors for Cancer Immunotherapy

Wednesday, 28th September - 17:40 - Nanomedecine for cancer diagnosis & therapy - Tower 24 - Room 103 - Oral presentation - Abstract ID: 20

Ms. Flavia Fontana [1], ***Dr. Mohammad-Ali Shahbazi*** [1], ***Dr. Dongfei Liu*** [1], ***Dr. Hongbo Zhang*** [1], ***Mr. Ermei Mäkilä*** [2], ***Prof. Jarno Salonen*** [2], ***Prof. Jouni Hirvonen*** [1], ***Dr. Helder A. Santos*** [1]

1. *University of Helsinki,* ***2.*** *University of Turku*

Introduction

Cancer is a very challenging disease and new approaches are sought to help in its treatment. Cancer immunotherapy involves the patient's own immune system in the fight against the tumors using antibodies directed to the so-called "check-point inhibitors" or by the administration of chimeric T-lymphocytes. It is envisaged that the administration of nanoparticles in cancer vaccines would lead to a decrease in the side effects of the drugs, helping targeted drug delivery, and increasing the efficacy of the therapy due to the adjuvant properties of the particles themselves [1].

Porous silicon (PSi) is a biocompatible porous material which presents adjuvant-like properties [2]. Acetylated dextran (AcDX) and spermine-modified AcDX (SpAcDX) are chemically-modified polymers derived from dextran, a FDA approved material, able to stimulate an immune response by the activation of Toll-like receptors [3].

Methods

Multistage nanovectors were prepared by nanoprecipitation during glass capillary microfluidics [4]. PSi nanoparticles were encapsulated in a polymeric matrix made of AcDEX. The immunostimulant properties of the nanoparticles were assessed in vitro on human immortalized immune cells and blood-derived monocytes.

Results

The developed nanosystems induced the expression of co-stimulatory factors (CD86) in, for example, peripheral blood monocytes (Fig.1).

Discussion

The multistage nanovectors, produced with different polymers promoted the expression of CD86 from human monocytes, KG 1, and BDCM (results not shown). Moreover, the system modified with membranes derived from cancer cells showed the highest levels of CDs stimulation, with a statistically significant difference compared to the particles with the polymer alone.

Conclusions

The developed systems promoted the expression of costimulatory factors (CD86) confirming their adjuvant properties that make them suitable platforms for further development as cancer vaccines.

References

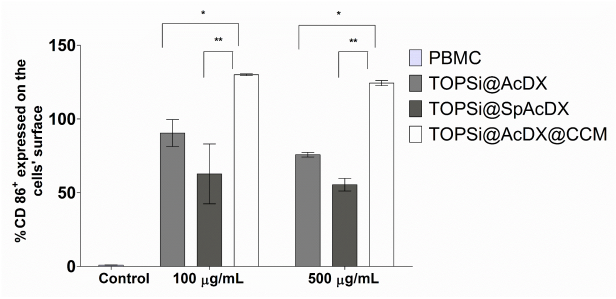
[1]D.J. Irvine,M.C. Hanson, 115 Chem Rev (2015), 11109-46.

[2]M.A. Shahbazi,T.D. Fernandez, Biomaterials 35 (2014), 9224-35.

[3]L. Cui,J.A. Cohen, Bioconjug Chem 22 (2011), 949-57.

[4]D. Liu,S. Cito, Adv Mater 27 (2015), 2298-304.

Acknowledgements
Financial support from the Academy of Finland (Grants No. 252215 and 281300), the University of Helsinki Research Funds, the Biocentrum Helsinki, and the European Research Council (FP/2007-2013, Grant 310892) is greatly acknowledged.



Cd 86 expression on pbmc.png

Thrombolytic therapy based on P-selectin targeted polymer nanoparticles

Wednesday, 28th September - 16:00 - Targeted drug delivery and Nanocarriers - Nano-Imaging for diagnosis, therapy and delivery - Tower 24 - Room 105 - Oral presentation - Abstract ID: 378

Ms. Maya Juenet [1], ***Ms. Rachida Aid-launais*** [1], ***Dr. Véronique Ollivier*** [1], ***Ms. Alice Berger*** [1], ***Mr. BO LI*** [1], ***Dr. Didier Letourneur*** [1], ***Dr. Cédric Chauvierre*** [1]

1. *INSERM U1148*

Injection of tissue plasminogen activator (tPA) remains the standard treatment for thrombolysis. However, high doses have to be administered with the risk to cause hemorrhages [1]. In our work, tPA-loaded nanoparticles were investigated to decrease the injected amount while keeping the same efficiency by ensuring a specific delivery. For this purpose, we synthesized polymer nanoparticles (NPs) targeting P-selectin as this protein is indeed a biomarker of activated platelets composing the thrombus.

Fluorescent polysaccharide-poly(isobutylcyanoacrylate) nanoparticles were synthesized by redox radical emulsion polymerization. They were functionalized with fucoidan (Fuco-NPs) as this anionic polysaccharide shows a nanomolar affinity for P-selectin [2]. Carboxymethyl-dextran was used as control (Control-NPs). To validate the targeting strategy, an in vitro flow assay was set up. Human whole blood was injected into collagen coated micro-channels to induce platelets activation and aggregation. Adhesion of NPs onto aggregates in venous conditions was quantified by fluorescence microscopy. tPA (Actilyse®) was loaded onto NPs by adsorption. The drug-related activity of the suspensions was measured in vitro and used to normalize the doses. Finally, an in vivo study was performed in a mouse model of mesenteric thrombosis induced by iron chloride.

Both NPs were similar in size, surface charge and fluorescence intensity. Fuco-NPs bound significantly more than Control-NPs to platelet aggregates confirming the potential of Fucoidan-functionalized NPs for targeting activated platelets (Figure A). The preclinical study showed that only Fuco-NPs-tPA were able to induce complete thrombolysis (4 mice out of 11) at a dose four times lower than the recommended one for murine models (Figure B). This result reinforced the choice of an active targeting approach, compare to other delivery strategies [3].

Fuco-NPs were efficient as carriers for thrombolysis enabling to decrease the injected tPA dose and therefore the risk of undesirable side effects. Although the variability between individuals is high in the developed model, this study underlines the great potential of targeted nanomedicine to fight thrombotic diseases.

[1] J. Alvazer-Sabin et al. Lancet Neurology 12(7) (2013)

[2] L. Bachelet et al. Biochimica et Biophysica Acta 1790(2) (2009)

[3] M. Varna et al. Future Science OA 1(4) (2015)

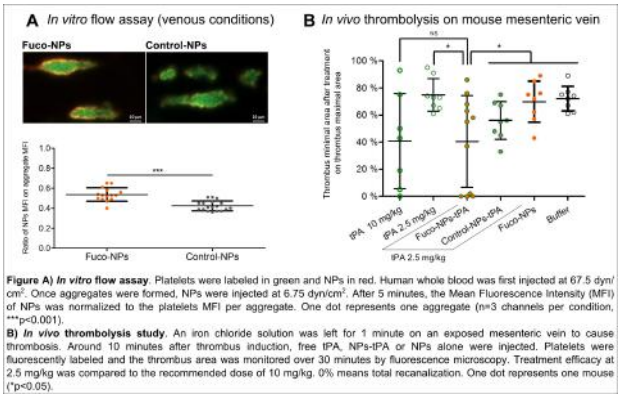


Figure A) *In vitro* flow assay. Platelets were labeled in green and NPs in red. Human whole blood was first injected at 67.5 dyn/cm^2. Once aggregates were formed, NPs were injected at 6.75 dyn/cm^2. After 5 minutes, the Mean Fluorescence Intensity (MFI) of NPs was normalized to the platelets MFI per aggregate. One dot represents one aggregate (n=3 channels per condition, ***p<0.001).

B) *In vivo* thrombolysis study. An iron chloride solution was left for 1 minute on an exposed mesenteric vein to cause thrombosis. Around 10 minutes after thrombus induction, free tPA, NPs-tPA or NPs alone were injected. Platelets were fluorescently labeled and the thrombus area was monitored over 30 minutes by fluorescence microscopy. Treatment efficacy at 2.5 mg/kg was compared to the recommended dose of 10 mg/kg. 0% means total recanalization. One dot represents one mouse (*p<0.05).

Mjueneticonan2016.png

Uptake and intracellular localization of engineered gold nanoparticles in A549 cells.

Wednesday, 28th September - 16:25 - Targeted drug delivery and Nanocarriers - Nano-Imaging for diagnosis, therapy and delivery - Tower 24 - Room 105 - Oral presentation - Abstract ID: 380

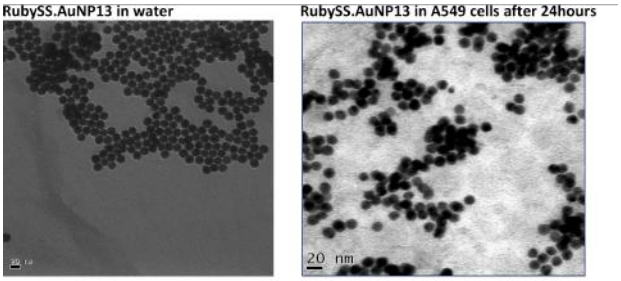
Ms. Abiola Dosumu [1], ***Ms. Shani Osborne*** [1], ***Prof. Zoe Pikramenou*** [1], ***Dr. Nik Hodges*** [1]

1. *University of Birmingham*

Engineered gold nanoparticles (AuNPs) possess unique physiochemical properties such as large surface area to mass ratio. Their relative ease of synthesis including incorporation of transition metal-ligands and other labelling agents produces functionalized AuNPs with potential applications for therapeutic and diagnostic medicine. Prior to clinical applications, an understanding of uptake, localization and cellular fate in relation to surface properties is critical.

In the current study we synthesised 13 nM AuNPs coated with a luminescent ruthenium metal complex to give rise to RubpySS.AuNP13. A non-toxic concentration (0.9 nM, MTT assay) was chosen to study particle uptake in A549 human lung carcinoma cells. Cells were incubated with RubpySS.AuNP13 (2-72 hours), uptake and co-localisation with different cellular compartments was studied using fluorescent probes (mitochondria, endoplasmic reticulum and golgi) and GFP-tagged organelle markers (early endosome, lysosome and autophagosome) by confocal microscopy supplemented by transmission electron microscopy (TEM). The size of particles in cells as quantified by TEM was in agreement with characterisation data of particles in solution (DLS and TEM) and particle size remained unchanged after up to 72 hours incubation indicating that RubpySS.AuNP13 does not aggregate upon cellular uptake. Particle number increased over time and this was confirmed by quantitative analysis (ImageJ) and further supported by TEM.

Both confocal and TEM images suggested the involvement of the endo-lysosomal pathway as a route of cellular uptake of RubySS.AuNP13 in A549 cells. RubySS.AuNP13 showed no significant co-localisation with markers for mitochondria, endoplasmic reticulum and the golgi and this was confirmed by TEM. In contrast, we observed particles in the early endosomes at early time points (4 hours), after 12-72 hours particles were observed to accumulate in a time-dependent manner in both the lysosomal and autophagosomal compartments of the cell, again this was confirmed by TEM. Also evident was a change in diffuse cytoplasmic LC3 staining to more punctuate staining consistent with the formation of active autophagosomes. In conclusion, we report that a major route of cellular uptake of RubySS.AuNP13 is by the endo-lysosomal pathway and furthermore accumulation of particles into autophagosome may have important implications for their cellular fate and toxicity.



TEM images of RubySS.AuNp13 in water and after 24hours incubation in A549 cells shows particles remains the same upon cellular uptake.

Rubyss bbb.png

Development of polymer microcapsules functionalized with fucoidan to target P-selectin under arterial flow conditions

Wednesday, 28th September - 16:50 - Targeted drug delivery and Nanocarriers - Nano-Imaging for diagnosis, therapy and delivery - Tower 24 - Room 105 - Oral presentation - Abstract ID: 352

Mr. BO LI [1], ***Ms. Maya Juenet*** [1], ***Dr. Véronique Ollivier*** [1], ***Ms. Rachida Aid-launais*** [1], ***Dr. Didier Letourneur*** [1], ***Dr. Cédric Chauvierre*** [1]

1. *INSERM U1148*

Introduction

The lack of technology, which enables early detection and predicts rupture risk of aneurysms hampers their diagnosis and treatments. P-selectin expressed by activated platelets and endothelial cells is a marker of biologically active arterial thrombi. Our group previously demonstrated that fucoidan-based systems could target P-selectin to detect platelet and endothelial activation in vivo. Therein, microcapsules (MCs) made of polycyanoacrylate and polysaccharide and functionalized with fucoidan (Fuco-MCs) were designed as new carriers to target P-selectin. Flow channels were used to visualize and quantify the affinity of Fuco-MCs for P-selectin and for human activated platelet aggregates expressing P-selectin in arterial flow conditions.

Methods

Microcapsules (MCs) were synthesized by anionic emulsion polymerization in alkaline condition. Vena8 Fluoro+ chambers were coated with a solution either with recombinant P-, E- or L-selectin. Some channels had different doses of P-selectin to confirm that the affinity of Fuco-MCs for P-selectin was depended on the concentration of P-selectin. A suspension of MCs or Fuco-MCs was passed through the channels (shear stress 1,500 s-1). In the second experiment, whole human blood was firstly injected into collagen coated channels to induce platelets activation and aggregation, subsequently MCs or Fuco-MCs were injected in the channels, visualized and quantified under fluorescence microscopy.

Results

In this work we, developed microcapsules with a new alkaline solvent emulsion-evaporation polymerization process, confirmed their hydrodynamic diameter distribution, the microcapsule structure and the presence of the targeting agent, fucoidan, at the surface. Fuco-MCs bound significantly more to the P-selectin coating than MCs, and results indicated a dose-dependent effect while increasing the concentration of P-selectin. Furthermore, their adhesion to L- and E-selectins was significantly lower than to P-selectin. Moreover, only Fuco-MCs accumulated at the surface of platelet aggregates.

Conclusion

This work gave the first evidence that microcapsules can be obtained according to one-step polymerization process of isobutyl cyanoacrylate in alkaline conditions. Subsequently, the microcapsule surface is functionalized by fucoidan, which gives microcapsules the property of specificity to P-selectin under blood flow conditions. Meanwhile, organic core is considered as a potential loading space of probe and/or hydrophobic drug for the diagnosis and/or the treatment of vascular diseases overexpressing P-selectin.

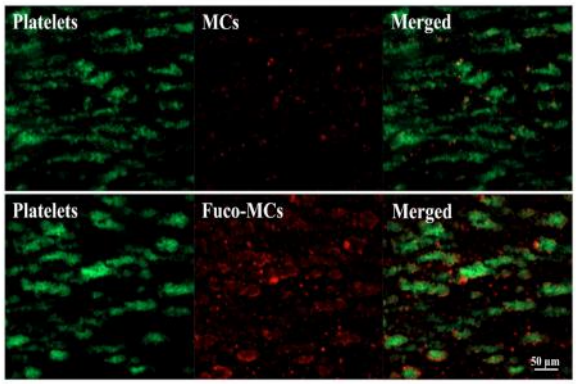


Figure. Microcapsule adhesion over platelet aggregates. Whole blood labeled with Dioc6 was injected onto channels coated with collagen at 50 µg/mL. Under arterial flow conditions, platelets adhered onto the channel wall forming aggregates expressing P-selectin (Left) and FITC-MCs or FITC-Fuco-MCs were then injected for 5 minutes (Middle panel). Channels were washed with 0.9% NaCl. Microcapsules and aggregates co-localization was assessed by merged fluorescence microscopy (Right).

Bo li - iconan 2016.jpg

Ligand Tethered Gold Nanoparticles against Untamed and Drug Resistant Leishmania Donovani

Wednesday, 28th September - 17:15 - Targeted drug delivery and Nanocarriers - Nano-Imaging for diagnosis, therapy and delivery - Tower 24 - Room 105 - Oral presentation - Abstract ID: 326

Prof. Arup Mukherjee [1], ***Mr. Asim Halder*** [1], ***Dr. Suvadra Das*** [1]

1. *University of Calcutta*

Introduction: Gold nanoparticles (Aunp) are useful tools in chemical biology interfaces. Engineered Aunps in sub-100 nm ranges are often successful in cancer chemotherapy and infections control. We have designed quasi-spherical Aunps and functionalized them further in lactoferrin conjugation reactions. New generation ligand tethered nanoparticles were experimented successfully against Leishmania donovani parasites.

Methods: Green synthesis for highly mono-dispersed Aunps was carried out using gallic acid. Reductive formation of Aunps at 40C under sonication was monitored in Uv-Vis spectrophotometer and the particles recovered by centrifugation 16000 rpm, 30 min. Aunps were re-suspended in water and conjugated in 1,2 ethylenediamine. Human lactoferrin (Lf) was further functionalized onto Aunps in EDC-NHS reactions. Plasmonic Lf-Aunps uptake in mouse peritoneal macrophage was monitored and leishmanicidal efficacy against axenic and macrophage infested L. donovini AG 83 strains were studied in depth.

Results and Discussion: Aunps formation was monitored in Uv-Vis spectrophotometer (Fig 1). Aunps appeared quasi-spherical with average TEM size near 7.6 nm. The zeta potential was recorded at – 24.2 mv and PDI was 0.21. Lf-Aunps observed relative increase in size (Fig 1E) and the zeta potential was -18.2 mv. Diffractions indexed in XRD studies were at (1 1 1), (2 0 0), (2 2 0) and (3 1 1). Aunp and Lf-Aunp mass compositions were analyzed in ICP-EES. Lf-Aunps were exceedingly effective against both axenic and the macrophage resident amastigotes. Comparative evaluations under culture conditions confirmed Lf-Aunp IC50 4.1±1 µM and 2.0±0.3 µM against L. donovini AG83 axenic and macrophage infested forms. Lf-Aunps were equally effective against sodium stibogluconate resistant strains but were refractory in paromomycin resistant organisms. Nanoparticle remarkable efficacy against macrophage infested leishmania was reasoned due to targeted transport, rapid uptake and ani-oxidant potentials.

Figure 1: Nanoparticles synthesis and efficacy against L. donovini AG83.

Nanoparticle(Aunp) TEM,A; SAED,B; Plasmonic response in time scale, C; In different gallic acid concentrations, D; Lf-Aunp TEM studies,E; Lf-Aunp TEM uptake in macrophages

(30 min exposure),F.

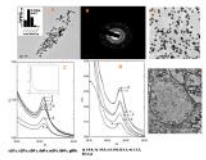


Figure 1.png

Harnessing human blood to examine bio-nano interactions at the cellular level

Wednesday, 28th September - 17:40 - Targeted drug delivery and Nanocarriers - Nano-Imaging for diagnosis, therapy and delivery - Tower 24 - Room 105 - Oral presentation - Abstract ID: 187

Mr. Joshua J Glass [1], ***Ms. Liyu Chen*** [2], ***Dr. Michael Whittaker*** [3], ***Ms. Sarah Mann*** [3], ***Prof. Edmund Crampin*** [1], ***Dr. John Quinn*** [3], ***Ms. Ewa Czuba*** [3], ***Dr. Kristofer Thurecht*** [2], ***Dr. Georgina Such*** [1], ***Prof. Thomas Davis*** [3], ***Dr. Angus Johnston*** [3], ***Dr. Robert De Rose*** [1], ***Prof. Stephen Kent*** [1]

1. *University of Melbourne,* ***2.*** *University of Queensland,* ***3.*** *Monash Institute of Pharmaceutical Sciences*

Aim: The capacity to predict interactions between nanomaterials and primary human cells will benefit the rational design of novel nano medicines. However, these fundamental relationships have not been adequately defined. By examining various nanoparticle systems with fresh human blood cells, we aim to establish principles of bio-nano interactions and generate predictive relationships that can inform the next generation of nanomedicines.

Methods: Four separate nanoparticle technologies were used to generate particles with varying charge and surface chemistries. Each particle was incubated with fresh healthy human blood (to study cell association/uptake) or purified cells (to study immune activation) at 37˚C and 4˚C and analysed by flow cytometry.

Results: The properties of nanoparticles significantly impact immunological outcomes, including immunoactivation and leukocyte uptake.

a.Hyperbranched polymers (HBPs) differentially activate primary human blood dendritic cells based on charge. HBP of 8 nm diameter were prepared and tested under endotoxin-free conditions. Cationic (+16 mV) HBPs activated myeloid dendritic cells (DC; 51.7 ± 4.3% IL-8+) but not plasmacytoid DCs, while neutral and anionic (-15 mV) HBPs were not immunostimulatory to either subset.

b.Charge dictates HBP association with different immune cell subsets. Cationic HBPs associated with most cell types (monocytes, granulocytes, DCs and B cells), while at 37˚C anionic HBPs preferentially associated with cells specialized for pathogen clearance and processing.

c.Nanoparticles engineered with terminal disulphides displayed enhanced association with human blood components, particularly phagocytic cells and platelets, over control nanoparticles.

d.Altering the chemical composition of nanoparticle surfaces can reduce phagocytic clearance. Linear poly(ethylene)glycol surfaces reduced phagocytic uptake compared with brushed poly(ethylene)glycol of the same molecular weight.

Conclusion/Future: We are in the process of creating a nanoparticle ‘characteristic matrix’ to examine the effect of single nanoparticle characteristics on immunoactivation and blood interaction profiles. We will subsequently apply mathematical modelling principles to generate predictive models of bio-nano interactions.

Injectable thermoresponsive magnetic hydrogel composite incorporating iron oxide and hydroxyapatite nanoparticles for bone tissue engineering

Wednesday, 28th September - 16:00 - Targeted drug delivery and Nanocarriers - Tissue engineering and regenerative nanomedicine - Tower 24 - Room 107 - Oral presentation - Abstract ID: 505

Dr. Padmalosini Muthukumaran [1], ***Prof. Seeram Ramakrishna*** [2], ***Prof. Balázs Gulyás*** [1], ***Prof. Raju V. Ramanujan*** [3], ***Prof. Dinesh Kumar Srinivasan*** [1]

1. *Lee Kong Chian School of Medicine, Nanyang Technological University,* ***2.*** *Centre for Nanofibers and Nanotechnology, National University of Singapore,* ***3.*** *School of Materials Science and Engineering, Nanyang Technological University*

Introduction: Injectable bone cements (IBC) are liquid at normal conditions, but when injected into bone defect, they will stabilize and mold to the defect shape and aids in bone regeneration. Commercially available IBC are based on polymethylmethacrylate, calcium phosphate or calcium sulfate. These IBC have several drawbacks such as low radiopacity, lack of bioactivity, heat induced necrosis of surrounding tissue, poor mechanical strength etc. The objective of this study is to fabricate a novel thermosensitive injectable bone cement with incorporation of super paramagnetic iron oxide nanoparticles and nanohydroxyapatite (nHA) and test their suitability of bone healing.

Methods: Composite hydrogel was prepared by homogenization of Poly (N-isopropyl acrylamide)-co-acrylic acid (pNIPAM-co-AA) polymer solution with citrate modified magnetic iron oxide nanoparticles (MNP, 15-20 nm), nHA (200 nm) and strontium ranelate (SR). The obtained hydrogel was characterized for its structural, thermal and mechanical properties. Human mesenchymal stem cells (hMSC) were grown on the composite hydrogels in vitro and the structure, proliferation, osteogenic differentiation and mineralization ability of the cells on the hydrogel was analysed using MTS, alkaline phosphatase (ALP) assays, alizarin red staining and immunocytochemistry.

Results: The hydrogel composite comprised of a microporous structure with uniform distribution of the nanoparticles. LCST of the composite was observed as 35oC with 80.25% water content. FT-IR analysis showed characteristic peaks for amide, acid, cyanide, phosphate and carbonate groups. The onset of thermal degradation was observed at 144oC with residual weight of 16% at 900oC. The cells grown on hydrogel composite showed significantly higher proliferation and ALP activity ($p<0.01$). Further, the quantification of ARS staining revealed significantly higher mineralization ($p<0.01$) in the composite hydrogel. Immunohistochemistry studies showed that the cells expressed both CD90 and osteocalcin, indicating that the cells are differentiating towards osteogenic lineage.

Discussion: The results suggest that the fabricated composite hydrogel could serve as injectable bone cements for non-invasive or minimally invasive defect filling in bone tissue engineering. The hydrogel is also capable of inducing mesenchymal stem cell differentiation towards osteogenic lineage.

Acknowledgement: This work was supported by MOE AcRF Tier 1 research grant (1T1-01/15) awarded to Asst. Prof. Dinesh Kumar Srinivasan, LKCMedicine, NTU.

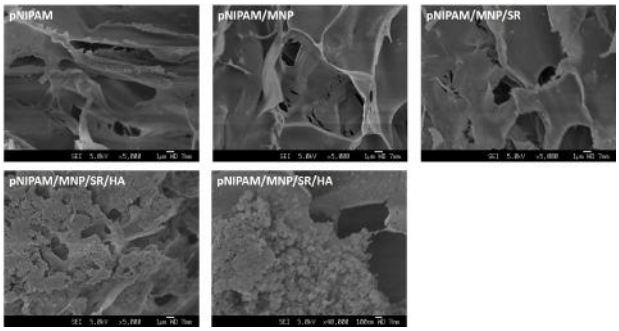


Figure structure of hydrogel.jpg

Biogenic Gold Nanoparticles for Complete Recovery of Dermal Burn Wounds

Wednesday, 28th September - 16:25 - Targeted drug delivery and Nanocarriers - Tissue engineering and regenerative nanomedicine - Tower 24 - Room 107 - Oral presentation - Abstract ID: 277

Dr. Suvadra Das [1], ***Mr. Asim Halder*** [1], ***Dr. Partha Roy*** [2], ***Ms. Anwesha Banerjee*** [3], ***Mr. Durbadal Ojha*** [3], ***Mr. Saptarshi Mandal*** [1], ***Dr. Debprasad Chattopadhyay*** [3], ***Prof. Arup Mukherjee*** [1]

1. *University of Calcutta,* ***2.*** *ADAMAS University,* ***3.*** *ICMR Virus Unit, I.D. and B.G. Hospital*

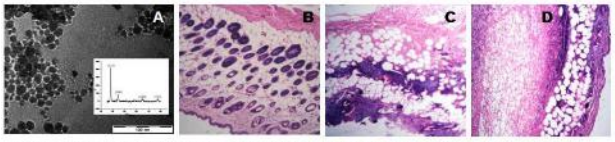
Introduction: Despite developments in regenerative medicine areas, the skin burn recovery remained a burdensome problem in critical care. The aim of this study was to arrive at safe flavonoid anchored gold nanoparticles and investigating them in dermal burn wound animal models.

Methods: Flavonolignan Silymarin is a proclaimed antioxidant and immunostimulant available from Silybum marianum. One pot bio-catalytic technique was applied for silymarin conjugated gold nanoparticles synthesis. Course of the reaction was followed systematically in LCMS and the appearance of gold plasmonic response was recorded in time scale. Silymarin conjugated gold nanoparticles (SmAunp) were characterized in DLS, XRD, FTIR, TEM, FESEM and ICPMS. Burn wound experiments were run in mice as per the guidelines of IAEC, University of Calcutta (Registration No. 506/01/a/CPCSEA, 2009-2010) approval No 506/01/a/CPCSEA/CUTech03. Experimental burn wound was inflicted under ether anaesthesia and the animals in group of 10 were treated with 5 µl SmAunp in water (600 µg/ml), standard silver sulfadiazine or saline control solutions. Dorsal skin burn was inflicted with 10 mm circular brass probe, 20 sec exposure at 100 0C. Recovery or not was recorded over a fourteen days treatment period. Histopathology and tissue biochemistry were recorded in each group intermittently and at the end of the experiment.

Results: Molecular bio-organic conjugation was confirmed in final product LCMS and in FTIR studies. Particle plasmon response was recorded at 540 nm and the average size in TEM was observed at 12.07 nm. SAED pattern and XRD observations proved fcc crystalline structure for the metallic gold. SmAunp exhibited excellent healing properties like wound closure, granulation, and vascularisation in comparison to standard drug. Histological analysis showed significant epithelialization in wounds treated with SmAunp.

Discussion: Wound healing properties of bio safe gold nanoparticles are reported for the first time. SmAunp exerted appropriate cytokines regulation and it is likely that the stem cells in the remaining hair follicular bulge were activated due to SmAunp. Favourable antioxidant properties of SmAunps were another factor for accelerated recovery.

Acknowledgement: Financial support to Dr. Suvadra Das from University Grants Commission (Sanction no. F.15-1/2015-16/PDFWM-2015-17-WES-33815(SA-II)), Government of India is thankfully acknowledged.



(A) TEM image of SmAunp (inset XRD spectrum of SmAunp). Histological features of partial thickness burn wounds on day 14 after burn injury (hematoxylin & eosin stain), (B) normal skin, (C) untreated wound, (D) SmAunp treated wound.

Tem xrd studies and histological features.jpg

WORKSHOP: Different approaches for the formation of synthetic hydrogels based on hybrid physically-chemically cross-linked networks

Wednesday, 28th September - 16:50 - Targeted drug delivery and Nanocarriers - Tissue engineering and regenerative nanomedicine - Tower 24 - Room 107 - Workshop presentation - Abstract ID: 413

Dr. Maxime Grillaud [1], ***Mr. Maarten Bakker*** [1], ***Dr. Patricia Dankers*** [1]

1. *Eindhoven University of Technology*

Many polymers with different types of cross-linking have been designed to develop various hydrogels for industrial and fundamental applications. In particular, they have attracted significant interest in the field of regenerative medicine, as drug delivery vehicles or scaffolds for tissue engineering. However, the main challenging limitation for their extensive use is their low mechanical stability. Many recent efforts have been made to overcome this burden, in particular by the elaboration of double network (DN) hydrogels. Despite the promising results of the few existing physically-chemically cross-linked DN hydrogels, the preparation procedures require long photopolymerization time and toxic precursor materials. Moreover, only the comparison of the mechanical properties between the hybrid DN hydrogel made in very specific conditions with the corresponding two separate networks is studied. The fundamental understanding of the combination of two different cross-links for a rational design of new hybrid DN hydrogels is a challenge.

Herein, we provide synthetic strategies to tailor in-situ formation of DN hydrogels based on physically-chemically cross-linked networks. Three approaches to combine catalyst-free click reaction and hydrogen bonding for in-situ hydrogel formation will be described (see Figure). More in detail, the covalent cross-linking consists of the click reaction between azides and cyclooctyne derivatives, referred to as strain-promoted azide-alkyne coupling (SPAAC), and the non-covalent cross-linking is based on supramolecular interactions via a four-fold hydrogen bonding motif, the ureido-pyrimidinone (UPy) unit.

The mechanical properties of these new hydrogels are studied by rheology and their morphology are analyzed by confocal microscopy. A detailed comparison of the hydrogel properties is discussed according to the approach selected and the ratio of the two types of cross-linking. In particular, this work shows the benefits to associate the dynamicity afforded by supramolecular interactions for gel recovering with the strength provided by the covalent cross-links for enhanced material stiffness. We propose that this work offers both a novel versatile platform for the easy preparation of DN hydrogels under physiological conditions with tunable properties, and one more step in the fundamental understanding of structure-property relationship of physically-chemically cross-linked hydrogels.

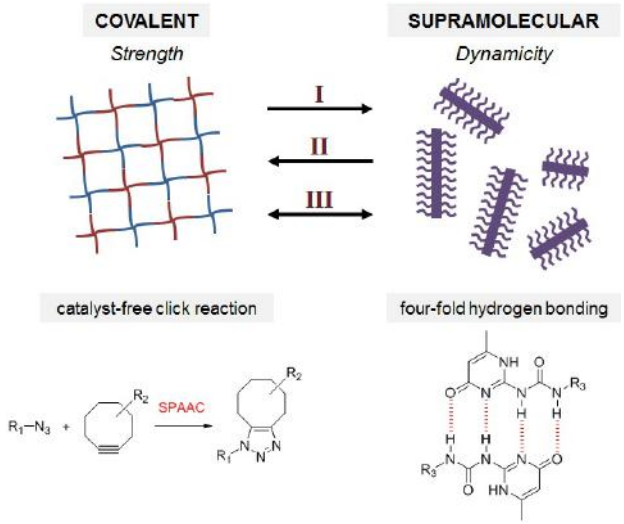


Figure abstract.jpg

The Drive to Master the Foundation Principles of Nanoscale interactions with living Organisms

Thursday, 29th September - 09:00 - Plenary Speeches - Amphitheatre 25 - Oral presentation - Abstract ID: 565

Prof. Kenneth Dawson [1]

1. *Centre for BioNano Interactions, School of Chemistry, University College Dublin*

The nanoscale is unique in biology, and our capacity to engineer on that scale will be transformative. Thus, the endogenous (intrinsic) machinery of biology is defined and operates on the nanoscale. Typical biomolecules and assemblies that are actively transported around organisms by specific motors and drivers are between 5-80nm. This means that nanoparticles are also actively (using the energy of the cell) transported around cells and biological barriers all unlike small molecules which passively partition into biological compartments (cells, organs etc). Secondly, the power of being able to communicate with, and use those endogenous mechanisms of biology is potentially transformative in practical terms. That is an enduring fact that renders our effort to draw the power of nano to the challenge of medicine. However, these could remain only words, or broad ideas in our generation, if we do not learn how to engage this enormous potential to interact with the machinery of organisms. There are
deep challenges. First of all, the complexity of the interaction is remarkable, much more so than for small cell and molecules, or large particles. Capturing this new capacity for benefit of human society will require dedication and commitment, indeed an exceptional generational effort, rather than peripheral or aspirational research. Both the potential, and the challenge, of this field may have been underestimated, but now now we have faced the need to invest in
guiding principles and laws governing the whole arena, the true picture is unfolding quickly. We are optimistic. We discuss progress being made in understanding how interactions between nanoscale objects and living organisms occur, and their governing principles. [2,3] We argue that the future lies in pressing forward to develop a truly microscopic (molecular scale) understanding between the nanoscale and living organisms. [4]

References

1. Proceedings of the National Academy of Sciences 104 (7), 2050-2055 (2007).
2. Nature nanotechnology 7 (12), 779-786 (2012)
3. Nature nanotechnology 7 (1), 62-68 (2012)
4. Nature nanotechnology 10 (5), 472-479 (2015)

Biocompatible Metal Organic Frameworks in Nanomedicine

Thursday, 29th September - 09:40 - Plenary Speeches - Amphitheatre 25 - Oral presentation - Abstract ID: 30

Dr. christian serre [1]

1. *ENS-ESPCI-PSL-CNRS*

Metal Organic Frameworks (MOFs) are a recent class of tuneable porous hybrid solids based on metal sub-units connected to organic complexing molecules (carboxylates, phosphonates, azolates…) delimiting an ordered porous network. Their unprecedented structural and chemical diversity is of potential interest for many applications such as gas storage, separation or catalysis, among others.(1) Recently, some of us demonstrated their high potential in biomedicine for drug delivery, release of biologically active gases or imaging.(2) This led in most cases to very high drug loading capacities and a prolonged release using a wide range of active drugs with different polarities and sizes.(3) We will report here the recent progresses in this field from the synthesis, degradability, toxicity and biodistribution of nanoparticles of biocompatible MOFs,(4) to their encapsulation, release and in vitro or in vivo properties.(5-10)

References

1-Themed issue: Metal-organic frameworks, Chem. Soc. Rev., 2014

2 M. Gimenez et al., Coord. Chem. Rev., 2015

3- (a) P. Horcajada et al. Angew. Chem., Int. Ed. 2006, 45, 5974

4- Baati, T.; et al. , Nat. Mater., 2010, 9, 172

5- T. Chalati, et al., Nanomedecine, 2011, 6(10), 1657

6- J.E. Eubank et al. APL Mater., 2014, 2, 124112

7- Agostoni, V et al.. Adv. Healthcare. Mater. 2013, 2(12), 1630-1637.

8- (a) E. Bellido et al. Langmuir, 2014, 30(20), 5911-5920; (b) E. Bellido et al. Adv. Health. Mater, 2015 4(8), 1246-1257

9- V. Agostoni et al. Scientific Reports, 2015, 5, 7925

10- Cooper, L. et al. Chem. Comm. 2015, 51(27), 5848-5851

Dendrimers as tools towards nanomedicine

Thursday, 29th September - 10:45 - Plenary Speeches - Amphitheatre 25 - Oral presentation - Abstract ID: 250

Dr. Anne-Marie Caminade [1]

1. *LCC-CNRS Toulouse*

Dendrimers [1] are hyperbranched and nanosized macromolecules, synthesized step-by-step in an iterative fashion to ensure a perfect control of all their structural parameters (chemical composition, size, weight, etc.), and which may have outstanding properties. [2] Most of these properties of dendrimers depend on their terminal functions, but surprisingly, the internal structure may also play a key role, especially when considering the biological properties. [3]

We synthesize phosphorus-containing dendrimers (i.e. dendrimers having a phosphorus atom at all the branching points), and we can play with both their internal structure [4] and the type of their terminal functions. These dendrimers, when specifically functionalized (see Figure), display many biological properties ranging from transfection to immuno-stimulation, [5] from antivirals to anti-prion, anti-inflammatory [5, 6, 7] and anti-cancer [8] agents. These biological properties of phosphorus-containing dendrimers will be emphasized.

References

[1] Dendrimers. Towards Catalytic, Material and Biomedical Uses. Caminade A.M., Turrin C.O., Laurent R., Ouali A., Delavaux-Nicot B, Eds., John Wiley & Sons, Chichester (UK) 2011.

[2] Caminade A.M., Ouali A., Laurent R., Turrin C.O., Majoral J.P., Chem. Soc. Rev. 2015, 44, 3890.

[3] Caminade A.M., Fruchon S., Turrin C.O., Poupot M., Ouali A., Maraval A., Garzoni M., Maly M., Furer V., Kovalenko V., Majoral J.P., Pavan G.M., Poupot R. Nature Comm. 2015, 6, 7722.

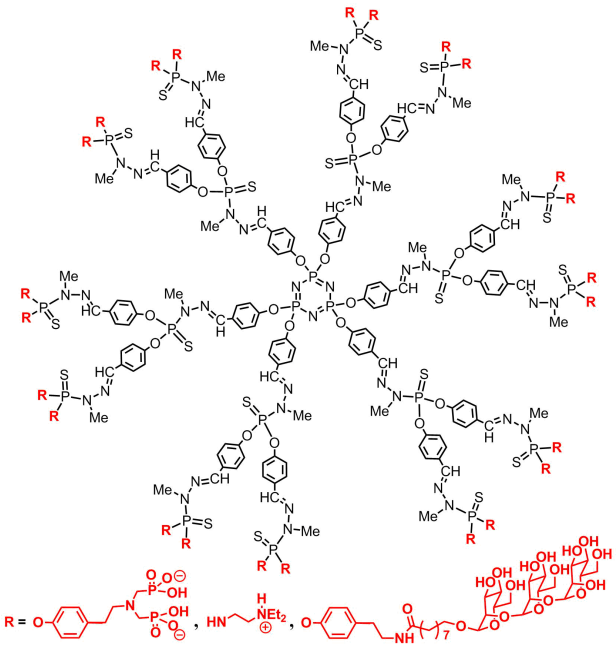
[4] Galliot C., Larré C., Caminade A.M., Majoral J.P., Science 1997, 277, 1981.

[5] Hayder M., Poupot M., Baron M., Nigon D., Turrin C.O., Caminade A.M., Majoral J.P., Eisenberg R.A., Fournié J.J., Cantagrel A., Poupot R., Davignon J.L., Science Transl. Med. 2011, 3, 81ra35.

[6] Blattes E., Vercellone A., Eutamène H., Turrin C.O., Théodorou V., Neyrolles O., Majoral J.P., Caminade A.M., Prandi J., Nigou J., Puzo G., Proc. Nat. Acad. Sci. USA 2013, 110, 8795.

[7] Le Dall J., Fruchon S., Garzoni M., Pavan G.M., Caminade A.M., Turrin C.O., Blanzat M., Poupot R. Nanoscale 2015, 7, 17672.

[8] El Brahmi N., Mignani S.M., Caron J., El Kazzouli S., Bousmina M.M., Caminade A.M., Cresteil T., Majoral J.P. Nanoscale 2015, 7, 3915.



Dendrimer bio.png

From Nano Shape & Self Recognition to Flexibility in Cancer Treatment and Differentiation

Thursday, 29th September - 11:25 - Plenary Speeches - Amphitheatre 25 - Oral presentation - Abstract ID: 527

Prof. dennis discher [1]

1. *University of Pennsylvania*

From viruses to tissue matrices, biology is filled with remarkable polymeric structures that motivate mimicry with goals of both clarifying and exploiting biological principles. Filamentous viruses inspired our development and computations of worm-like polymer micelles – 'filomicelles' – that persist in the circulation and deliver even better than spheres [1]. However, particles of any type interact with innate immune molecules and also phagocytes while nearby 'Self' cells are spared due to a polypeptide that limits phagocytic clearance [2]. The phagocyte's cytoskeleton forcibly drives the decision downstream of adhesion, proving analogous to how matrix elasticity directs stem cell fate [3, 4].

[1] Y. Geng, P. Dalhaimer, S. Cai, R. Tsai, M. Tewari, T. Minko, and D.E. Discher. Shape effects of filaments versus spherical particles in flow and drug delivery. Nature Nanotechnology (2007) 2: 249-255.

[2] P.L. Rodriguez, T. Harada, D.A. Christian, D.A. Pantano, R.K. Tsai, and D.E. Discher. Minimal 'Self' peptides that inhibit phagocytic clearance and enhance delivery of nanoparticles. Science (2013) 339: 971-975.

[3] A. Engler, S. Sen, H.L. Sweeey, and D.E. Discher. Matrix elasticity directs stem cell lineage specification. Cell (2006) 126: 677-689.

[4] J. Swift, I.L. Ivanovska, … and D.E. Discher. Nuclear Lamin-A Scales with Tissue Stiffness and Enhances Matrix-directed Differentiation. Science (2013) 341: 1240104-1 to 15.

Magnetic force-based skeletal muscle tissue engineering for in vitro drug testing

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 64

Dr. Akira Ito [1], ***Mr. Kazushi Ikeda*** [1], ***Mr. Ryusuke Imada*** [1], ***Dr. Masanori Sato*** [1], ***Dr. Yoshinori Kawabe*** [1], ***Prof. Masamichi Kamihira*** [1]

1. *Kyushu University*

Introduction: Skeletal muscle tissue engineering holds great promise for pharmacological studies. In vitro culture systems are available for drug discovery for patients with injured, diseased and age-related muscle. For drug testing, conventional 2D cell culture systems are based on formation of myotubes on tissue culture dishes. However, the 2D cell culture systems have limitations to mimic in vivo skeletal muscle functions mainly due to lacking the architecture of native muscles. One of the most important characteristics of skeletal muscle is contractile force generation ability, and tissue-engineered skeletal muscle constructs should mimic the architecture of native muscles and reproduce contractile force generation. In the present study, we demonstrate an in vitro system for drug testing using tissue-engineered skeletal muscle constructs. Methods: We have developed a fabrication method of functional skeletal muscle tissue constructs by using a magnetic force-based tissue engineering technique, in which myoblasts were labeled with magnetite nanoparticles, and assembled by magnetic field to form a cell-dense and aligned skeletal muscle-like structure. Small-molecular drugs are used as a model drug. Results and Discussion: In response to small-molecular drugs, myotube differentiation of myoblasts were promoted in 2D cell culture. However, the levels of contractile force generation of tissue-engineered skeletal muscle constructs treated with small-molecular drugs were not consistent with those of myotube differentiation in 2D cell culture. On the other hand, there was a high correlation between sarcomere formation as well as contractile activity in two-dimensional cell culture and contractile force generation of tissue-engineered skeletal muscle constructs. These observations indicate that the contractility data is indispensable for in vitro drug screening.

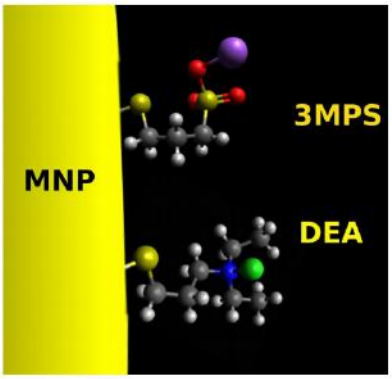
Characterization of Noble Metal Nanoparticles functionalized by molecule-capping method with mixed organic ligands carried out by SR-XPS and SERS

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 69

Ms. Laura Carlini [1], ***Prof. Chiara Battocchio*** [1], ***Prof. Paolo Postorino*** [2], ***Ms. Claudia Fasolato*** [2], ***Dr. Ilaria Fratoddi*** [2], ***Dr. Iole Venditti*** [2], ***Ms. Giovanna Testa*** [2], ***Dr. Fabio Sciubba*** [2]

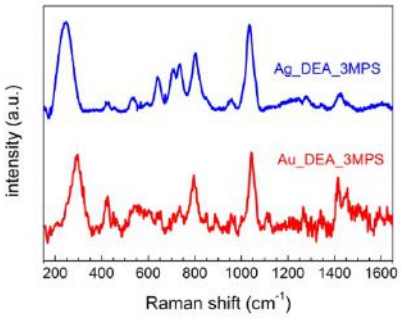
1. *Roma Tre University,* ***2.*** *Sapienza University of Rome*

This work is focused on the study of the structural, electronic and morphological properties of Noble Metal Nanoparticles (MNP-s) for innovative applications in nano-medicine and nano-biotechnology.A key point for the technological development of nano-structured materials and practical biomedical devices based on MNP-s is the achievement of a fine control of the stability and toxicity of the system.The chemical functionalization of the MNP-s can be done by means of capping metallic clusters with appropriate organic ligands.The molecule-capping method provide a reliable control of particle composition, shape and size distribution making the MNP-s suitable for active purposes in catalysis, nanoelectronics, sensing and bioanalysis.In this communication, we present MNP-s (Au and Ag) functionalized with mixed organic ligands (DEA and 3MPS) prepared with different metal/thiol stoichiometric ratios.The molecular overlayer has been selected on purpose for the biomedical applications, as well as to stabilize the nanoparticles.The changes in the stoichiometric ratio between metal and different capping agents can influence the chemical properties of ligands functional groups and the dimension of the functionalized MNP-s.Moreover, the biocompatibility of the system depends strictly on the charge and thickness of capping molecules layer.Therefore, the study of the interaction at the interface between the organic ligand and MNP-s at atomic level is the first step for the fabrication and developing of very suitable nanomaterials for diagnostic and therapy.The characterization of the nano-systems was carried out by means of Synchrotron Radiation induced X-ray Photoelectron Spectroscopy (SR-XPS) and Self Enhanced Raman Spectroscopy (SERS).SR-XPS provides information on the local bonding environment of a given species and it has been demonstrated to be a unique tool for investigating the nature of the interaction at the capping agent/metal nanoparticle interface, as well as the chemical structure of MNPs surface.In SERS, the Raman intensity diffused by molecules close to a nano-curved metallic surface is highly enhanced by the the localized surface plasmon resonance (LSPR), allowing the spectroscopical investigation of molecular monolayers.In conclusion, we compared the semi-quantitative SR-XPS and SERS analysis to obtain a better understanding of the system exploring the potential synergy between different techniques in order to give new insights in the field of nanomaterials.



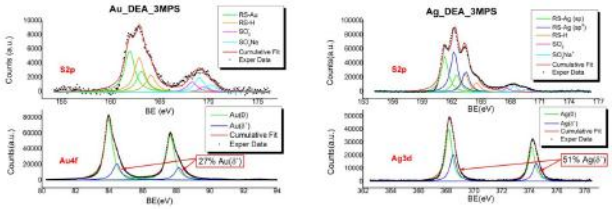
Noble Metal Nanoparticle functionalized by sodium 3-mercapto-1-propansulfonate (3MPS) and 2-diethylaminoethanethiol hydrochloride (DEA).

Functionalized mnp.jpg



SERS spectra of functionalized MNPs.

Sers spectra.jpg



SR-XPS spectra of S2p and metal (Au4f, Ag3d) core levels for **gold** and **silver** nanoparticles functionalized by DEA and 3MPS.

Sr-xps spectra.jpg

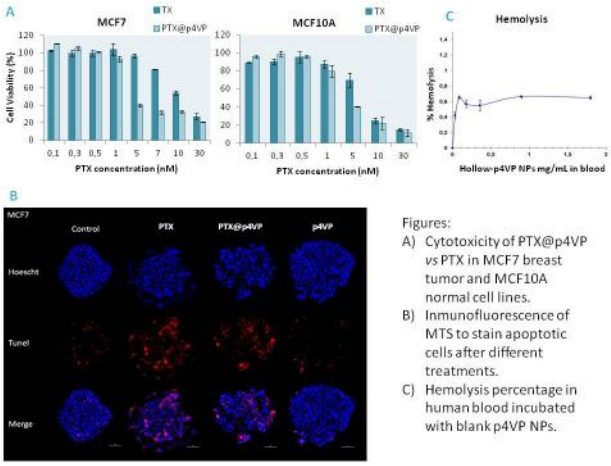
Incorporation of paclitaxel into hollow-p4VP nanoparticles to improve breast cancer chemotherapy

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 74

Ms. Maria del Carmen Leiva Arrabal [1], ***Ms. Julia Jiménez-López*** [1], ***Dr. Rafael Contreras-Caceres*** [2], ***Ms. Laura Cabeza*** [1], ***Dr. Gloria Perazzoli*** [1], ***Dr. Raúl Ortiz*** [3], ***Prof. Consolación Melguizo*** [1], ***Prof. Juan Manuel López-romero*** [2], ***Prof. Jose Prados*** [1]

1. *Institute of Biopathology and Regenerative Medicine (IBIMER), Center of Biomedical Research (CIBM), University of Granada, Granada, Spain; Biosanitary Institute of Granada (ibs.GRANADA), Andalusian Health Service (SAS), University of Granada, Spain.,* ***2.*** *Department of Organic Chemistry, University of Málaga, Málaga, Spain.,* ***3.*** *Institute of Biopathology and Regenerative Medicine (IBIMER), Center of Biomedical Research (CIBM), University of Granada, Granada, Spain; Department of Health Sciences, University of Jaén, Jaén, Spain.*

Introduction: Paclitaxel (PTX) is being currently used in the treatment of breast cancer, though it presents some drawbacks as low solubility, no tumor specificity and high toxicity. PTX loading into nanoparticles (NPs) may overcome those limitations, avoiding the use of toxic Cremophor EL in the formulation, and improving treatment efficiency. In this work, we have developed hollow poly(4-vinylpyridine) nanoparticles (p4VP) for PTX delivery, and their biocompatibility and antitumor activity against breast cancer has been in vitro studied.
Methods: Firstly, biocompatibility of p4VP was analyzed by a hemolysis assay with human blood. Sulphorrodamine B method was used to compare PTX and PTX-loaded p4VP (PTX@p4VP) cytotoxicity on human breast MCF7 cancer cells and MCF-10A normal cell line. Moreover, their activity against multicellular tumor spheroids (MTS) formed from MCF7 cells, was studied by tracking their volume every 2 days, and by immunostaining with TUNEL to compare cell death after different treatments.
Results: Hemolysis assay with human blood proved no toxicity of p4VP. In addition, cell death was improved by the use of PTX@p4VP in both cell lines, whereas the effect was more evident on tumor cells (2.9-fold reduction of PTX IC50 vs 1.7-fold). Furthermore, MTS treated with PTX@p4VP showed a significant volume reduction than those treated with PTX. However, both treatments achieved a reduction in MTS volume in comparison to controls and blank p4VP. Immunofluorescence corroborated those data by staining death cells on the surface of MTS treated with PTX and PTX@p4VP.
Discussion: According to those results, p4VP present good biocompatibility that make them suitable for in vivo drug delivery. They are also able to enhance PTX antitumor effect against breast cancer cells and breast MTS, which resemble tumor mass. In the light of our findings, PTX@p4VP represent a promising delivery system for the improvement of chemotherapy involved in the treatment of breast tumor.



Figures:

A) Cytotoxicity of PTX@p4VP *vs* PTX in MCF7 breast tumor and MCF10A normal cell lines.
B) Inmunofluorescence of MTS to stain apoptotic cells after different treatments.
C) Hemolysis percentage in human blood incubated with blank p4VP NPs.

P4vp nps with ptx against breast cancer.jpg

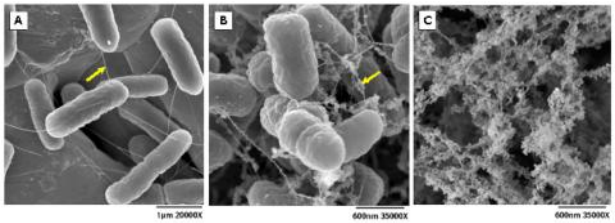
Tannin-chitosan composite nanoparticles as potencial nanomedicine to prevent urinary tract infections

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 77

Dr. Sergio Madrigal-Carballo [1], ***Ms. Emilia Alfaro-Viquez*** [2], ***Dr. Christian Krueger*** [2], ***Prof. Jess Reed*** [2]

1. *National Center for Biotechnological Innovations (CENIBiot), CeNAT-CONARE,* ***2.*** *Reed Research Group, University of Wisconsin-Madison*

Introduction. Naturally occurring renewable resources that inhibit microbial adhesion are important alternatives to antibiotics and may help stem the alarming increase in drug resistant bacteria. Bacterial invasion of gut epithelial cells (enterocytes) provides a mechanism by which pathogens are protected from complements, antibodies, and other immune defense molecules, which in turn, allows the pathogens to colonize and persist in the gut. We have invented novel composite biomaterial comprising formulations of tannin, extracted from cranberries (fruit, juice or press cake), and chitosan, manufactured from shrimp shells. The composites are versatile and can be formulated into useful biomaterials, such as nanoparticles, 3-dimensional foams, and films. Methods. Chitosan binds to negatively charged tannins by an electrostatic interaction driven by its positively charged amino group. This interaction allows developing stable hybrid nanoparticles via ionotropic gelation with tripolyphosphate (TPP), suitable as a therapeutic controlled release system for urinary tract infections. We study the effect of cranberry tannin-chitosan composite nanoparticles on the invasion of Caco-2 cells by Uropathogenic Escherichia coli (UPEC) strain 5011, supplied by the UW-University Hospital (Madison, WI, USA). Results. Results of a dose-response experiment indicate the cranberry tannin-chitosan composite nanoparticles significantly reduced invasion of Caco-2 cells by extra-intestinal pathogenic E. coli (ExPEC). The nanoparticles significantly inhibited the ability of the pathogen to invade the Caco-2 cells by 40%, 80% and 96% at a total polyphenolic concentration of 0.2 µg, 0.5µg and 0.75µg GAE/ml (respectively). Discussion. Figure shows scanning electron micrographs exploring the effect of tannin-chitosan composite nanoparticles on ExPEC surface structures that are involved in cell adhesion and subsequent inhibition of invasion of intestinal epithelial cells in vitro. When the pathogen was exposed to the tannin-chitosan composite nanoparticle material (Fig. C), extensive coating and cross-linking on multiple bacteria was seen (Fig. B), compared to control (Fig. A). Results suggest the cranberry tannin-chitosan composite nanoparticles physically coat the surface virulence factors of ExPEC, which in turn prevents invasion of the intestinal epithelial cell.



Tannin-chitosan nanoparticles interactions with expec.png

Lung cancer: a new approach to paclitaxel treatment using PLGA nanocarriers

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 90

Ms. Julia Jiménez-López [1], ***Ms. Maria del Carmen Leiva Arrabal*** [1], ***Dr. Mazen El-hammadi*** [2], ***Ms. Laura Cabeza*** [1], ***Dr. Gloria Perazzoli*** [1], ***Dr. Lucía Martín-Banderas*** [2], ***Dr. Raúl Ortiz*** [3], ***Prof. Jose Prados*** [1], ***Prof. Consolación Melguizo*** [1]

1. *Institute of Biopathology and Regenerative Medicine (IBIMER), Center of Biomedical Research (CIBM), University of Granada, Granada, Spain; Biosanitary Institute of Granada (ibs.GRANADA), Andalusian Health Service (SAS), University of Granada, Spain.,* ***2.*** *Pharmacy and Pharmaceutical Technology, University of Sevilla,* ***3.*** *Institute of Biopathology and Regenerative Medicine (IBIMER), Center of Biomedical Research (CIBM), University of Granada, Granada, Spain; Department of Health Sciences, University of Jaén, Jaén, Spain.*

Introduction

Lung cancer is the type of cancer with the highest mortality rate worldwide. Paclitaxel (PTX), an antimitotic drug, is widely administered for the treatment of choice for this type of cancer and other solid tumours. However, PTX has many limitations and is associated with many undesirable effects like neutropenia, neurotoxicity or hypersensitivity reactions. In this work a new type of polymeric nanoparticles of lactic and glycolic acid (PLGA) loaded with this drug for the treatment of lung cancer is proposed to solve these limitations.

Methods

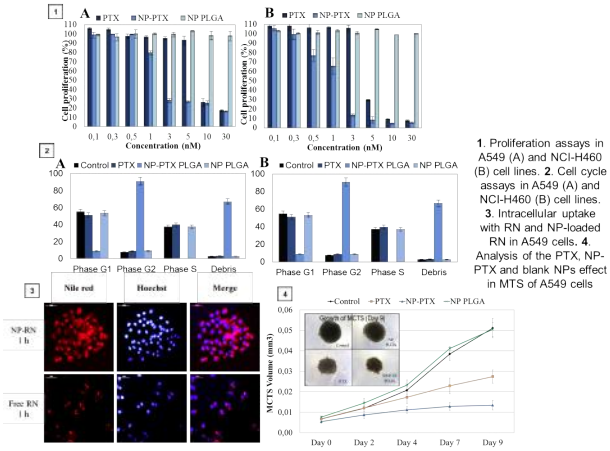
Proliferation assays with sulforhodamine B in two human lung tumor cell lines, A549 and NCI-H460, were performed in order to compare the effect of free PTX and NPs loaded with this drug (NP-PTX). Cytotoxicity studies with blank nanoparticles were performed. The mechanisms of action of PTX and NP-PTX were tested with cell cycle assays by flow cytometry. For cellular uptake studies with fluorescence microscopy, A549 was incubated with Nile red (NR) and NR-loaded NPs at different times. Finally, multicellular tumour spheroids (MTS) assays in A549 were performed and MTS were treated with PTX, NP-PTX and blank NPs at the IC50 dose of free PTX. MTS growth was monitored with imaging microscopy.

Results

The results obtained in proliferation assays showed significant dose reduction IC50 with NP-PTX up to 3.63 and 3.79 times lower in A549 and NCI-H460, compared to free PTX. No inhibition was observed in the growth of any cell lines with blank nanoparticles. The results showed enhanced cell cycle accumulation in G2 and sub-G1 phase in cells treated with NP-TX facing PTX-treated cells. Intracellular uptake results suggest that NP-PTX facilitate the incorporation of NR into the cell compared with free NR. Finally, the MTS assays showed a significant and greater reduction in the volume of spheroids treated with NP-PTX (73%) than treated with free PTX (46%). No significant difference was observed between MTS untreated and treated with blank NPs.

Discussion

In short, these promising in vitro results obtained suggest that PLGA NPs provide a potential strategy as a mechanism for PTX encapsulation and could allow increasing the therapeutic index of this drug.



1. Proliferation assays in A549 (A) and NCI-H460 (B) cell lines. **2**. Cell cycle assays in A549 (A) and NCI-H460 (B) cell lines. **3**. Intracellular uptake with RN and NP-loaded RN in A549 cells. **4**. Analysis of the PTX, NP-PTX and blank NPs effect in MTS of A549 cells

In vitro results of a polymeric nanoparticle loaded with paclitaxel for the treatment of lung cancer.png

Nanofibers Preparation by Free-Liquid Surface Electrospinning for Cartilage Tissue Engineering

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 95

Ms. Parinita Agrawal [1], ***Prof. Krishna Pramanik*** [1]

1. *National Institute of Technology, Rourkela, Odisha*

Cartilage degeneration due to aging like osteoarthritis, or developmental abnormalities and trauma causes severe damage to cartilage tissue. Adult cartilage tissue has limited self-repair capacity thus the deformity is generally not self-cured. Tissue engineering can provide cure to this problem by designing scaffold of appropriate composition to mimic the extra cellular matrix of the damaged tissue on which the stem cells could grow to form functional cartilage tissue. Electrospinning is the method for formation of nanoscale diameter fibrous matrix, which provides high surface area, beneficial for cell adhesion. Free-liquid surface electrospinning is an advanced method that provides higher productivity. In the present study, we explore electrospinning for fabricating scaffolds from chitosan (CS) blended with regenerated silk fibroin (SF). To improve the processing ability of the blend solution, poly(ethylene oxide) (PEO) was added to the blend which was then successfully electrospun. SF is a naturally occurring biodegradable fibrous protein having good mechanical properties and biocompatibility. CS has intrinsic antibacterial activity and biocompatibility that makes it suitable for the tissue engineering applications.

The morphology of nanofibrous matrix was characterized by scanning electron microscopy (SEM). Crystalline nature and hydrophilicity of the nanofibrous matrix was confirmed by X-ray diffractometry and contact angle analysis, respectively. The matrix was found to possess biodegradation rate and tensile strength adequate for cartilage tissue replacement. The electrospun nanofibers also supported growth and proliferation of mesenchymal stem cells (MSCs) as observed by field emission SEM and MTT studies. Histological and fluorescence microscopic analysis confirmed the differentiation of MSCs to chondrocytes, on the nanofibrous matrix. The confirmatory study by quantitative-PCR validated the potential of the prepared nanofibrous matrix for cartilage tissue engineering applications.

Keywords: Nanofiber, electrospinning, tissue engineering, chitosan, silk fibroin, poly(ethylene oxide)

Evaluation of chloroaluminium phthalocyanine-loaded magnetic nanoemulsion as drug delivery device to treat glioblastoma using hyperthermia and photodynamic therapy

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 101

Dr. Leonardo Barcelos de Paula [1], Prof. Fernando Lucas Primo [2], Prof. Marcelo Rodrigues Pinto [3], Prof. Paulo Cesar Morais [4], Prof. Antonio Claudio Tedesco [1]

1. *Department of Chemistry - University of Sao Paulo - USP - Ribeirao Preto/SP,* ***2.*** *Department of Bioprocess and Biotechnology - Univ. Estadual Paulista Júlio de Mesquita Filho - UNESP - Araraquara/SP,* ***3.*** *University of Uberaba - Uberaba/MG,* ***4.*** *College of Chemistry and Chemical Engineering - Anhui University - Hefei*

Introduction: Brain tumors known as glioma are the most common neoplasms found in the central nervous system of adults. In a brain cancer tumor cancer stem cells (CSCs) represent a small fraction of the total cancer cell population. Studies have suggested similarities of CD133+ CSC with the proneural subtype and of CD133 CSC with the mesenchymal subtype. Methods: We developed two samples magnetic nanoemulsions (MNEs) loaded with magnetic nanoparticles (MNPs) plus photoactivated drugs, chloroaluminium phthalocyanine (ClAlPc), (0.15×1016 or 1.50×1016 MNP/mL), were obtained through a spontaneous emulsification process based on oil-in-water mixtures. All the cell lines (human mesenchymal stem cells derived from bone marrow - BM-MSC and the human glioma - U87MG and T98G) were cultived in alpha-MEM, DMEM-low and 10% fetal bovine serum at 37 ºC and 5% CO2, respectively. The AC magnetic field during the hyperthermia (HPT) studies operates at 1 MHz with 40 Oe magnetic field amplitude. Photodynamic therapy (PDT) protocol used a typical diode laser set at 670 nm and operates with 100, 200, and 700 mJ/cm2 fluency. For different treatments the cell viability was assessed by the MTT test. Results and Discussion: Particle size analysis showed that all colloidal formulations produced in the present study and containing ClAlPc were on the nanoscale size, with typical mean diameter around 200 nm and size distribution value of 0.25. Cytotoxicity assay was conducted with the MNE/ClAlPc samples at the two distinct MNP concentrations while keeping fixed the ClAlPc concentration (0.5 μmol/L). Synergism of the combined HPT and PDT treatments, revealing an increase of up to 60-70% in cell death. Conclusion: From the present study we found that the HPT treatment reduces the cell viability in about 15%, regardless the magnetic nanoparticle content within the MNE. However, a superior reduction in cell viability of about 70% was found while combining the HPT and PDT treatments. Moreover, confocal microscopy images clearly indicated the cytoplasm localization and active site of the as-prepared drug delivery device. Therefore, we envisage that the combined treatment of HPT and PDT represents a promising paradigm for brain cancer intervention, such as glioblastoma.

Synthesis, research and functionalization of hybrid contrast agents based on gadolinium doped magnetite

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 136

Mrs. Iana Tcareva [1], ***Prof. Alexander Majouga*** [2], ***Prof. Alexander Savchenko*** [1], ***Mr. Maksim Abakumov*** [3], ***Mr. Igor Shchetinin*** [1]

1. *National University of Science and Technology, MISiS,* ***2.*** *Lomonosov Moscow State University,* ***3.*** *The Pirogov Russian National Research Medical University (RNRMU)*

Magnetic Resonance Tomography, MRI, is one of the most used instruments for non-invasive clinical diagnostics. In contrast to radiological investigations, MRI has no danger of radiation exposure to produce images using radio frequency electromagnetic radiation with very low energy. The most researches in this area are focused on the development of contrast agents, that can provide a clearer distinction between healthy and diseased tissue. The majority of contrast agents are magnetic nanoparticles (MNP).

MNPs of iron oxide are used for the diagnostics of many diseases such as cardiovascular, neurological and cancer tumors. Liver and prostate cancer are the most prevalent of malignant tumors. Currently a priority researches of contrast agents for MRI are hybrid contrast agents, providing comprehensive diagnostic information about the dynamics of the disease progression. Gadolinium chelates and magnetite are the most appropriate T1 and T2 contrast agents, respectively, but these compounds can provide toxic effect on healthy cells. One of the ways to prevent toxicity is the creation of hybrid contrast agents based on gadolinium doped magnetite.

In this work gadolinium doped magnetite nanoparticles were prepared by two techniques: thermal decomposition of iron and gadolinium acetylacetonates in diphenyl ether and hydrolysis of Fe^{3+} и Gd^{3+} salts mixture in the presence of polyethyleneimine, followed by addition of hydrazine hydrate and microwave irradiation. These nanoparticles are designed to be used as a hybrid contrast agents for hepatocellular and prostate carcinomas visualization.

Obtained nanoparticles were investigated by methods of transmission electron microscopy, X-ray diffraction, Mössbauer spectroscopy, VSM magnetometry and thermogravimetric analysis. Also the toxicity of nanoparticles and their T2-relaxation time were measured in vitro. In vivo examinations on mouse model of liver cancer were carried out.

The authors knowledge financial support from Ministry of Education and Science of the Russian Federation (14.607.21.0132, RFMEFI60715X0132).

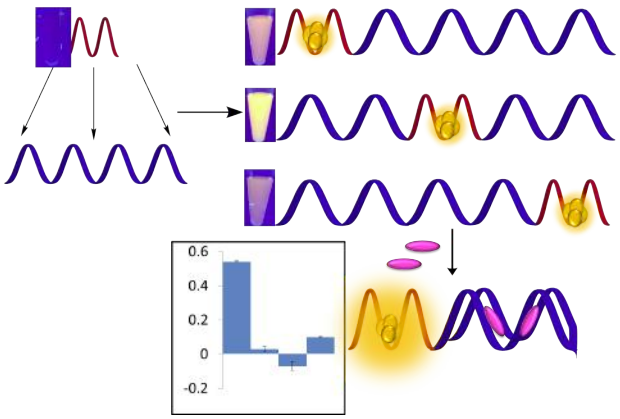
In-depth investigation on DNA-AgNCs designs for adenosine detection

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 184

Ms. Shi Ting Lee [1], ***Dr. Siu Yee New*** [1]

1. *The University of Nottingham Malaysia Campus*

Silver nanocluster (AgNCs) as like other metal nanoclusters has size of approaching Fermi wavelength of electron (2 nm). This nanomaterial no longer possesses surface plasmon resonance (SPR). Indeed, its energy structure is in a discrete level that similar to those of molecules, enable it to absorb intense light and emit strong fluorescence colour upon excited with UV light. Among stabilising ligands, DNA template has particularly gained more attention. Apart from its versatility, it may due to the feasibility to integrate DNA aptamer into the template. This aptamer functions exactly like antibody and binds specific to a target analyte. Hitherto, there is no detailed study about the impact of sequence template arrangement (i.e. AgNCs nucleation sequence and target recognizing sequence) on the sensing performance of AgNCs. In this work, we are investigating the effect of different combination between AgNCs nucleation sequence (NC) and adenosine aptamer (Apt A) onto adenosine detection. Three different designs have been formulated in order to systematically position AgNCs nucleation sequences and adenosine aptamer in the same template, namely Apt A 5, Apt A 3 and Apt A M with the last letter implies the position of AgNCs nucleation sequence at 5'-end, 3'-end and in the middle. Among these designs, only Apt A 5 design gives selective result against adenosine, with a 0.5-fold enhancement in the emission intensity observed. The linear range of detection is wide and across four order of magnitude, 1 – 2500 µM. We deduced that the fluorescent increment is correlated to the guanine rich bases in the aptamer sequence.



Graphic abstract - dna-agncs design to adenosine detection.png

Noncovalent Assembly of Carbon Nanotubes: Toward the Construction of Nanotube-Based Breast Cancer Therapy Nanovectors

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 204

Dr. Ayhan Unlu [1], Mr. Mehdi Partovi Meran [2], Ms. Bircan Dinc [3], Mr. Yasin Celikok [4], Prof. Isil Albeniz [5], Prof. Seniha Guner [2]

1. *Trakya University,* ***2.*** *Istanbul Technical University,* ***3.*** *Istanbul Kemerburgaz University,* ***4.*** *Yeni Yuzyil University,* ***5.*** *Istanbul University*

In recent years a wide range of different nanoscale drug delivery vectors have been evaluated. Among the numerous nanoscale systems, carbon nanotubes (CNTs) hold great potential for diverse applications and are becoming a viable component of biomedical science. Apart from their uses in the cellular imaging with diagnostic effects in nanomedicine, CNTs are promising drug carriers in the drug delivery systems for cancer therapies. One of the key advantages of CNTs in biomedical applications is that they can be easily internalized by cells and, therefore, can act as delivery vehicles for a variety of molecules relevant to therapy and diagnosis. In addition, the high aspect ratio of CNTs offer great advantages over existing delivery vectors, as the high surface area provides multiple attachment sites for drug targeting. The main obstacle for utilization of pristine CNTs are their scarce solubility in any aqueous environment due to the graphitic nature of their sidewalls. To overcome this problem, CNT surfaces should be modified by noncovalent coating or by covalent functionalization approaches. In this study, we describe a non-covalent Single-Walled Carbon Nanotubes (SWCNT)–doxorubicin supramolecular complex that can be developed for cancer therapy. In light of the unique properties of SWCNT and the hydrophilic and flexible ability of PEG chains, we linked PEG chains and pyrene molecules, which can strongly attach to SWCNT surface via π-π stacking. Different monocarboxylic acid-terminated PEGs were synthesized and subsequently PEG-Pyrene was prepared via an esterification reaction of PEG-COOH with pyrene methanol. Finally, SWCNT and PEG-pyrene heterogeneous mixture was sonicated and stirred vigorously in THF to gain PEG modified SWCNT. We initially examined the cytotoxicity of different PEG-modified SWCNT samples in MDA-MB-231 breast cancer cell line and determine IC50 values for this type cells by using MTT cell proliferation assay in our study. The results indicated that PEG-modified SWCNTs have significantly lower cytotoxicity. At the final stage, the interaction between Doxorubicin and functionalized SWCNT was studied by monitoring the emission spectrum of doxorubicin by fluorescence spectrophotometry.

Molecular dynamics simulation of interaction of lysine dendrimer and Semax peptide

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 214

Mrs. Elena Popova [1], ***Prof. Igor Neelov*** [2], ***Mr. Victor Kuznetsov*** [1], ***Mr. Sergey Petunov*** [1], ***Mr. Angrey Radilov*** [1]

1. *Research Institute of hygiene, occupational pathology and human ecology,* ***2.*** *ITMO University*

Peptide drug delivery systems development is one of the most important tasks in modern pharmacology. Therapeutic peptides are efficient and have minimum side effects. Parenteral route of peptide administration is the most effective. The main problem in oral route is the action of proteolytic enzymes on peptides and their low mucosal permeability. It is proposed to use dendrimers to solve the problem. Dendrimer is a macromolecule with a symmetrical branched structure. It has stable charge and size. Dendrimers reduce toxic effect of peptides and ensure targeted delivery into cells (e.g., cancer). In some cases, dendrimers with drugs complexes have a much more pronounced therapeutic effect. The regulatory peptide Semax (Met-Glu-His-Phe-Pro-Gly-Pro) as a model one was chosen. It has an antioxidant, antihypoxic and neuroprotective effects. It is used for acute ischemic stroke prevention, traumatic brain injury, diseases of optic nerve and glaucoma optic neuropathy treatment, during rehabilitation after stroke.

Currently, method of molecular dynamics is the main method of polymer complexes and biopolymer systems modelling. The simulation was performed via this method for systems consisting of one lyzine dendrimer molecule (32 NH3+ groups), 8, 16 and 24 Semax molecules, water and chloride counterions in a cubic cell with periodic boundary conditions. Further energy minimizations and simulations using GROMACS 4.5.6 package and AMBER_99SB-ildn force fields were performed. The potential energy of this force field consists of valence bonds and angles deformation energy, angles of internal rotation, Van der Waals and electrostatic interactions. Complexes with peptides were formed in 30-60 ns. The equilibrium size and anisotropy of the complexes were close. Radial distribution of atom number shows that dendrimer was inside, and peptides mainly on the surface of complexes. Hydrogen bonds and ion pairs number of the third and the second complexes were less because of the less close contact between dendrimers and peptides. It is shown that dendrimer of third generation can attach about 22 Semax molecules.

Keywords: dendrimers, peptides, computer simulation, molecular dynamics method.

Salinomycin nanoparticles induce selective toxicity toward tumor cells rather than stroma in orthotopic model of pancreatic cancer

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 229

Dr. zahra Daman [1], ***Dr. Yuhua Wang*** [2], ***Prof. Leaf Huang*** [2]

1. *Department of Pharmaceutics, Faculty of Pharmacy, Tehran University of Medical Sciences, Tehran, Iran,* ***2.*** *Laboratory of drug targeting, Eshelman School of Pharmacy, University of North Carolina in Chapel Hill, NC, USA*

Introduction

Salinomycin (SAL), a selective inhibitor of cancer stem cells (CSCs), was recently recognized for its ability to inhibit proliferation and induce apoptosis in various tumors including highly chemo-resistant ones. The aim of this study was to deliver SAL to orthotopic model of pancreatic cancer by the aid of poly(lactic-co-glycolic acid) (PLGA) nanoparticles (NPs) and explore some mechanistic questions regarding this treatment.

Methods

SAL-loaded NPs were prepared and investigated in terms of pharmaceutical properties. MTT assay was used to study cell proliferation in AsPC-1-luc cells. In vivo antitumor study was performed in nude mice orthotopically implanted with AsPC-1-luc cells and treated by IV administration of saline and SAL NPs for control and test groups, respectively. Tumor growth was monitored weekly through in vivo imaging of luciferase activity. The collected tumors from SAL-treated and control groups were used for protein extraction and immunofluorescent staining of different markers.

Results

Nano formulation of SAL was prepared in suitable size and loading traits. SAL (3.5 mg/kg every other day) blocked tumor growth by 52% compared to the control group after nine injections. Western blotting of tumor protein extracts indicated that SAL treatment leads to up-regulation of E-cadherin and β-catenin expressions in AsPC-1 orthotopic tumor. Noteworthy, immunofluorescence staining of adjacent tumor sections demonstrated that treatment with SAL NPs leads to considerable apoptosis in the tumor cells rather than the stroma.

Discussion

Herein, we utilized PEG-PLGA as a biocompatible and biodegradable polymer to formulate SAL into polymeric NPs, so that it could be stably solubilized for further investigations. Regarding in vivo treatments, a 52% reduction in tumor size compared to the control group highlights SAL-loaded PLGA NPs as a promising system for pancreatic cancer treatment. The results of concurrent CD31/α-SMA staining and the TUNEL assay showed that the apoptotic pancreatic cancer cells were mainly distributed in the tumor nest, which confirms efficient intratumoral extravasation and penetration of SAL-loaded NPs. Our data suggested that SAL can harness epithelial-mesenchymal transition through increased co-expression of E-cadherin and its binding protein, β-catenin. However, more mechanistic research is needed before any translation of SAL-based treatments into clinics.

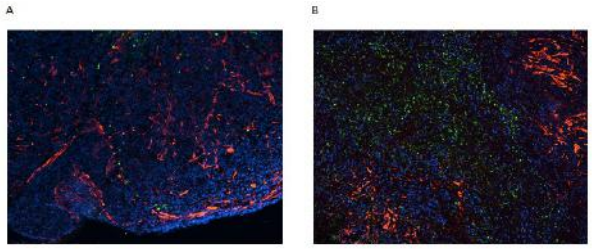


Fig iconan.jpg

Antibacterial effects of gold-chitosan nanocomposites on human macrophages infected by intracellular pathogenic bacteria

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 267

Dr. Gracia Mendoza [1], ***Ms. Anna Regiel-futyra*** [2], ***Dr. Vanesa Andreu*** [1], ***Dr. Víctor Sebastián*** [3], ***Dr. Agnieszka Kyzioł*** [2], ***Prof. Grażyna Stochel*** [2], ***Dr. Manuel Arruebo*** [3]

1. *University of Zaragoza,* ***2.*** *Jagiellionan University,* ***3.*** *Universidad de Zaragoza*

Introduction: Bacteria have developed resistances to antibiotics with the subsequent negative consequences in the treatment of different pathogenic infections. Nanomaterials have emerged as a potential bactericidal approach to avoid multidrug resistances. We previously demonstrated that gold-chitosan nanocomposites show bactericidal but not cytotoxic action at the doses tested (Regiel-Futyra et al., 2015). Here, we evaluated their action in a co-culture model of macrophages infected by different bacteria to closely mimic a real infection scenario.

Methods: Nanocomposites were prepared by a solvent evaporation method by using chitosan with medium average molecular weight and chitosan based gold nanoparticles dispersions with two different initial gold precursor concentrations, 1 and 2 mM. Then, gold nanoparticles and chitosan-gold nanocomposites were characterized by UV-Vis spectroscopy, transmission electron microscopy and Fourier Transform Infrared Spectra analysis. Cytotoxic effects on different cell lines and bacteria were evaluated separately (Regiel-Futyra et al., 2015). Cytotoxic effects of gold chitosan nanocomposites on macrophages infected with Gram positive (Staphylococcus aureus) and Gram negative bacteria (Escherichia coli) were evaluated and the antimicrobial activity was studied by the colony-forming unit assay.

Results: Gold chitosan nanocomposites exhibited potent antibacterial effects at the subcytotoxic concentrations assayed in both infection macrophage models being more accentuated in the Gram positive bacteria model in which one of our experimental groups exerted a complete bactericidal effect obtaining no bacteria colonies after treatment of the infected macrophages with the gold-chitosan based nanocomposites.

Discussion: The development of novel bactericidal strategies is imperative regarding the high prevalence of multidrug resistances arising in bacteria. By 2050 antimicrobial resistance would lead to 10 million people deaths every year and a reduction of 2% to 3.5% in Gross Domestic Product. Our study showed that at non cytotoxic concentrations, gold chitosan nanocomposites were able to kill both Gram positive and Gram negative bacteria in infection models in vitro without damaging host cells and avoiding the well-known cyto- and genotoxic effects of other metal nanomaterials such as silver and copper nanoparticles. This approach represents a potential basis in the design and development of bactericidal materials to efficiently halt the progression of bacteria colonization avoiding the growing problem of multidrug resistances.

Multifunctional polymer-modified liposomes that capture and neutralize toxic protein, histones

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 361

Mr. Hiroki Tsuchida [1], ***Dr. Hiroyuki Koide*** [1], ***Mr. Masahiko Nakamoto*** [2], ***Ms. Anna Okishima*** [3], ***Ms. Saki Ariizumi*** [3], ***Ms. Chiaki Kiyokawa*** [3], ***Dr. Tomohiro Asai*** [1], ***Prof. Yu Hoshino*** [2], ***Prof. Naoto Oku*** [1]

1. Department of Medical Biochemistry, Graduate School of Pharmaceutical Sciences, University of Shizuoka, 2. Department of Chemical Engineering, Kyushu University, 3. Department of Medical Biochemistry, School of Pharmaceutical Sciences, University of Shizuoka

Poly-N-isopropylacrylamide (pNIPAm) based polymer nanoparticles (NPs) with the capacity to recognize, capture and neutralize the target toxic peptide melittin in vivo by multiple weak interactions are of significant interest as “plastic antidotes”. The multipoint interactions are generated by N-tert-butylacrylamide (TBAm, a hydrophobic monomer) and acrylic acid (AAc, a negatively charged monomer). However, the NPs have low biocompatibilities and short circulation time. To improve these problems, we focused on modification of linear polymers prepared by reversible addition-fragmentation chain transfer (RAFT) polymerization of NIPAm, TBAm and AAc onto liposomal surface. Histones, major mediators of sepsis, were chosen as target proteins. The linear polymer library was prepared by changing the AAc ratio and polymer length. Liposomal sizes and zeta-potentials were not significantly different between before and after polymer modification onto the liposomal surface even in the increasing of AAc ratio and polymer length. The modification rates of polymers depend on AAc ratio and polymer length. The circulation times of polymer-modified liposomes were far longer than linear polymers. Interactions between polymer-modified liposomes and histones were monitored by quartz crystal microbalance (QCM). As a result, polymers including higher density of AAc enhanced affinity of liposomes to the histones. In addition, there is an optimal length to induce high affinity to the histones. We next demonstrated toxin neutralization effect of polymer-modified liposomes using mouse endothelial 2H-11 cells. The optimized polymer-modified liposomes inhibited histones cytotoxicity in vitro. These results indicate that the liposomes bound and neutralized target protein, histones. To confirm whether polymer-modified liposomes neutralize histones in vivo, optimized polymer-modified liposomes were intravenously injected into mice after the lethal injection of histones via tail vein. The polymer-modified liposomes improved survival rate of mice exposed to lethal dose of histones. Namely, polymer-modified liposomes neutralized histones in the bloodstream. The functional liposomes capable of capturing and neutralizing the target biomacromolecules can be used as new antidotes.

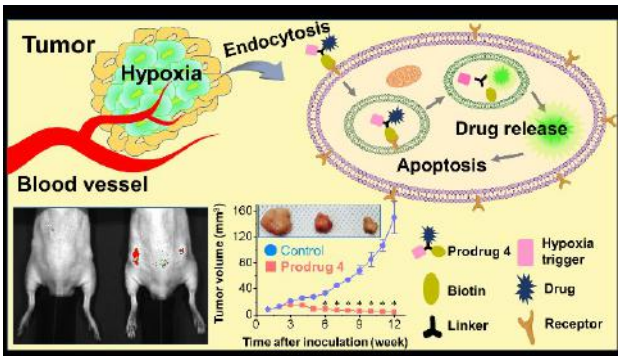
Hypoxia-directed and activated theranostic agent: Imaging and treatment of solid tumor

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 366

Ms. Hyun Min Kim [1], ***Prof. Eun-joong Kim*** [2], ***Mr. Hyunseung Lee*** [1], ***Prof. Jong Seung Kim*** [3], ***Dr. Kwan Soo Hong*** [1]

1. *Bioimaging Research Team, Korea Basic Science Institute,* ***2.*** *Department of Biomedical Engineering, School of medicine, Kyung Hee University,* ***3.*** *Department of Chemistry, Korea University*

Hypoxia, a distinguished feature of various solid tumors, has been considered as a key marker for tumor progression. Inadequate vasculature and high interstitial pressures result in relatively poor drug delivery to these tumors. Herein, we developed an antitumor theranostic agent, 4, which is activated in hypoxic conditions and can be used for the diagnosis and treatment of solid tumors. Compound 4, bearing biotin, a tumor-targeting unit, and SN38, an anticancer drug, proved to be an effective theranostic agent for solid tumors. SN38 plays a dual role: as an anticancer drug for therapy and as a fluorophore for diagnosis, thus avoids an extra fluorophore and limits cytotoxicity. Compound 4, activated in the hypoxic environment, showed high therapeutic activity in A549 and HeLa cells and spheroids. In vivo imaging of solid tumors confirmed the tumor-specific localization, deep tissue penetration and activation of 4, as well as the production of a strong anticancer effect through the inhibition of tumor growth in a xenograft mouse model validating it as a promising strategy for the treatment of solid tumors.



Hypoxia figure1.jpg

Chemically cross-linked silk fibroin hydrogel with enhanced elastic properties, biodegradability, and biocompatibility

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 368

Mr. minhee kim [1], ***Mr. seung hyun lee*** [1], ***Prof. Won Ho Park*** [1]

1. *Chungnam national university*

Introduction

In this study, the synthesis of SF hydrogel via a chemical cross-linking reaction using γ-ray irradiation was investigated and the resultant hydrogel was characterized. Two different hydrogels were investigated: SF P-gel induced by β-sheet structure and chemically cross-linked hydrogel prepared by γ-ray irradiation. The secondary structures and strengths of the gels were compared. Furthermore, the swelling behavior, biodegradation, and cell viability of human mesenchymal stem cells (hMSCs) on the SF hydrogel were evaluated.

Methods

Freshly regenerated SF solutions of various concentrations (2.3%–7.9%) were poured into a petri dish and irradiated with γ-rays from a Co-60 source. The radiation dose varied from 15–60 kGy and the dose rate was 15 kGy/h, at room temperature. The irradiated samples were cut into small pieces to compare with SF P-gel with regard to various properties.

Result and discussion

The radiation technique seems to be an excellent pathway for the preparation of hydrogels because polymer solutions undergo chemical cross-linking on irradiation to yield a hydrogel. By contrast, most chemical cross-linking reactions of SF can induce a modification of proteins, resulting in structural destabilization and inflammation by exposure to toxic organic solvents and cross-linking agents. In this study, the SF C-gel was prepared by γ-ray irradiation, which induced intermolecular cross-linking reactions. In order to investigate the effect of irradiation on the gelation of the SF solution, the solutions were γ-ray irradiated at various doses of 15, 30, 45, 60, 75, and 90 kGy. Regardless of the SF concentration and irradiation doses, SF hydrogel was formed immediately after irradiation, as shown in Figure 1A. However, as shown in Figure B, cracks occurred in the hydrogel at doses higher than 60 kGy. A suitable absorption dose for an SF solution was 60 kGy. Therefore, the experiment was performed using the dose of 60 kGy.

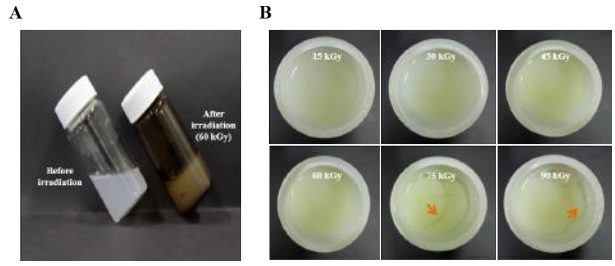

A
B
After irradiation (60 kGy)
Before irradiation
15 kGy
30 kGy
45 kGy
60 kGy
75 kGy
90 kGy

Figure.png

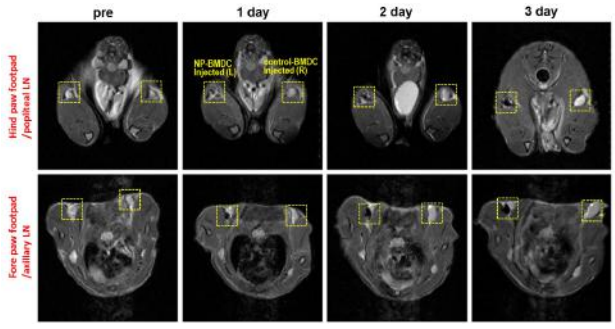
Non-invasive In Vivo Imaging and Tracking of Dendritic Cells Migration using MR/NIR Dual Modal Contrast Agent

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 373

Dr. Jee-Hyun Cho [1], ***Ms. Hyun Min Kim*** [2], ***Dr. Hye Sun Park*** [2], ***Dr. Kwan Soo Hong*** [2]

1. *Bioimaging Research Team, Korea Basic Institute,* ***2.*** *Bioimaging Research Team, Korea Basic Science Institute*

Dendritic cells (DCs) play a key role in immune system as potent antigen-presenting cells (APC), which are uniquely capable of initiating a primary immune response. After DCs capture and process antigen in peripheral tissues, they begin to mature and migrate to secondary lymphoid tissues, where they activate naïve T cells. Therefore, DC migration into the draining lymph nodes is critical for T cell priming. Here, we developed a novel magnetic resonance (MR)/near-infrared (NIR) imaging contrast agent, which was synthesized by a facile fluorescent dye coating on magnetic nanoparticles (MNP). The dye-coated MNP was used as a high-performance MR/NIR imaging contrast agent with a high T2 relaxivity (r2) value of approximately 308 mM-1s-1, and NIR fluorescence by biocompatible indocyanine green dye to support non-invasive in vivo tracking of DCs migration into lymph nodes. The impact of the dye-coated MNPs on DC viability and maturation was systematically investigated using MTT assays and FACS analysis. And the labeling effects of DCs with the dye-coated MNPs were investigated in changes on DC phenotype or maturation potential. Importantly, the migration of DCs via lymphatic drainage and homing into the lymph node were monitored through real-time NIR fluorescence and MR imaging. Taken together, this novel MR/NIR contrast agent has potential application as a non-invasive imaging agent for both inducing T cell priming and imaging of DC-based immunotherapy.



Abstract2 figure.jpg

Matryoshka-type enteric microparticles for the treatment of tuberculosis

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 376

Dr. Vanesa Andreu [1], ***Ms. Ane Larrea*** [1], ***Dr. Salvador Alfaro*** [2], ***Ms. Begoña Gracia*** [1], ***Dr. Gracia Mendoza*** [1], ***Dr. Víctor Sebastián*** [1], ***Dr. José Antonio Ainsa*** [1], ***Dr. Manuel Arruebo*** [1]

1. *University of Zaragoza,* ***2.*** *National Polytechnic Institute*

Introduction

The World Health Organization reported in 2015 that the tuberculosis still constitutes a major cause of morbidity and mortality worldwide. The treatment used nowadays presents different limitations such as non-localised delivery of drugs, high dose and large treatment duration as well as the side effects that it produces.

The aim of this work was to investigate a new polymeric drug-delivery system that may provide enhanced drug delivery at the site of infection, hence reducing drug associated side-effects and the probability of developing drug resistance. In that sense, matryoshka-type enteric microparticles containing rifampicin-loaded PLGA nanoparticles were developed as a potential effective oral therapeutic alternative to achieve appropriate intracellular concentrations in tuberculosis-infected alveolar macrophages.

Methods

Rifampicin-loaded PLGA nanoparticles (NPs) were prepared by the oil-in-water single-emulsion solvent-evaporation method and those NPs were encapsulated in turn within enteric monodisperse Eudragit-based-microcapsules using the double emulsification and evaporation method. The morphological characterization of micro- and nanoparticles has been carried out by several techniques (SEM, TEM, DLS, FT-IR).

In vitro antimycobacterial activity of rifampicin, unloaded-PLGA nanoparticles and rifampicin-loaded PLGA nanoparticles were determined on M. tuberculosis H37Rv at intracellular and extracellular pH. Also, in vitro permeability of these NPs through the intestinal epithelium was evaluated using an artificial monolayer of gastroinstestinal epithelial cells.

Results

The colloidal hydrodynamic sizes of rifampicin-PLGA NPs were 145.2 ± 31.2 nm. HPLC analysis indicated that the EE and DL achieved for these NPs were 5.4 ± 0.55 and 1.08 ± 0.11 w/w %, respectively. SEM morphological analysis of enteric Eudragit-microcapsules revealed a bipyramidal structure of micrometric size (6.1 ± 1.9 μm). The enteric coating remains unaltered under simulated gastric conditions and the inner antibiotic-loaded NPs were rapidly released under simulated intestinal conditions.

Antimicrobial activity of the free and the encapsulated rifampicin showed similar levels of bacterial eradication.

Discussion

An intracellular accumulation and slow antibiotic release can be achieved using orally administered enteric microparticles containing rifampicin-loaded PLGA NPs if they were able to extravasate through the intestinal epithelium into the blood stream with the sufficient blood circulation half-life to reach tuberculosis-infected alveolar macrophages.

Regulation of angiogenesis through the efficient delivery of microRNAs into endothelial cells using polyamine-coated carbon nanotubes

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 377

Dr. Andrea Masotti [1], ***Dr. Mark Miller*** [2], ***Ms. Antonella Celluzzi*** [1], ***Dr. Lorrain Rose*** [2], ***Dr. Federico Micciulla*** [3], ***Dr. Patrick Hadoke*** [2], ***Dr. Stefano Bellucci*** [3], ***Dr. Andrea Caporali*** [2]

1. *Ospedale Pediatrico Bambino Gesù,* ***2.*** *University of Edinburgh,* ***3.*** *INFN-Laboratori Nazionali di Frascati*

Introduction

MicroRNAs (miRNAs) directly regulate gene expression at a post-transcriptional level and represent an attractive therapeutic target for a wide range of diseases. Here, we report a novel strategy for delivering miRNAs to endothelial cells (ECs) to regulate angiogenesis, using polymer functionalized carbon nanotubes (CNTs).

Methods

CNTs were coated with two different polymers, polyethyleneimine (PEI) or polyamidoamine dendrimer (PAMAM), followed by conjugation of miR-503 oligonucleotides as recognized regulators of angiogenesis.

Results

We demonstrated a reduced toxicity for both polymer-coated CNTs, compared with pristine CNTs or polymers alone. Moreover, polymer-coated CNT stabilized miR-503 oligonucleotides and allowed their efficient delivery to ECs. The functionality of PAMAM-CNT-miR-503 complexes was further demonstrated in ECs through regulation of target genes, cell proliferation and angiogenic sprouting and furthermore, in a mouse model of angiogenesis.

Discussion

This comprehensive series of experiments demonstrates that the use of polyamine-functionalized CNTs to deliver miRNAs is a novel and effective means to regulate angiogenesis.

How to enhance the functionality of microencapsulated cells by using graphene oxide nanoparticles

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 383

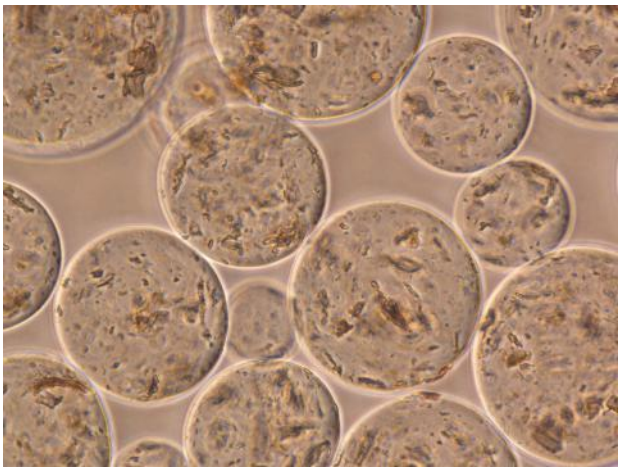
Dr. Laura Saenz del Burgo [1], ***Dr. Jesús Ciriza*** [1], ***Dr. Argia Acarregui*** [1], ***Mr. Haritz Gurruchaga*** [1], ***Prof. Rosa María Hernández*** [1], ***Dr. Gorka Orive*** [1], ***Prof. Jose Luis Pedraz*** [1]

1. *University of the Basque Country*

Introduction: Cell microencapsulation consists on the immobilization of cells within an alginate matrix surrounded by a semi-permeable membrane that allows the release of therapeutic drugs. This technology represents an opportunity to develop long-term de novo synthesized drug delivery systems for the treatment of chronic diseases. However, several challenges need to be overcome before it can be translated into the clinic. For instance, a higher viability of the cells would be highly desirable. Graphene oxide (GO) has shown to promote the adhesion and proliferation of several cell types. Thus, we fabricated hybrid alginate-GO capsules in order to improve the viability of erythropoietin (EPO) secreting C2C12 myoblasts.

Methods: Myoblasts were encapsulated into alginate or hybrid alginate-GO microcapsules. First, we studied the main physical parameters of the microcapsules. Next, early apoptosis and viability of the cells was quantified by flow cytometry. Also, the CCK8 assay was performed to analyze the metabolic activity and the cell membrane integrity was determined. Finally, EPO production was detected by Elisa. For in vivo studies, syngeneic C3H mice were implanted and blood samples were collected weekly for hematocrit determination.

Results and Discussion: We were able to produce microcapsules with GO in their core and proved that their physical characteristics were not substantially modified. Also, we determined that 25-50 μg/ml of GO is the most suitable concentration for enhancing the viability of encapsulated cells, since this rank provided lower cell death and better metabolic activity. However, even at the lowest GO concentration, the quantity of EPO detected on the medium was low, most likely due to its adsorption to the GO surface. Therefore, we tested if a protein pre-coating could attenuate the capacity of GO to adhere the therapeutic protein. Interestingly, serum-coating effectively avoided the adsorption of EPO to the nanoparticles and enhanced, even more, the viability, membrane integrity and drug release of encapsulated cells. Finally, myoblasts encapsulated in hybrid alginate-protein coated GO microcapsules showed to be functional in vivo as they increased hematocrit levels on mice. Therefore, these results provide another step for the future pre-clinical application of GO nanoparticles in combination with cell microencapsulation.



Microcapsules composed by 1.5 alginate and 200ug go 20x.jpg

Cellular internalization mechanisms of polyamine-coated carbon nanotubes

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 386

Ms. Antonella Celluzzi [1], ***Dr. Alessandro Paolini*** [1], ***Dr. Andrea Masotti*** [1]

1. *Ospedale Pediatrico Bambino Gesù*

Introduction

Carbon nanotubes (CNTs) are emerging as promising systems for transfection of drugs, small molecules and, recently, have been widely studied for the delivery of nucleic acids. In our previous paper, we reported the preparation of efficient delivery systems consisting of polymer-functionalized carbon nanotubes (CNTs). These systems efficiently delivered microRNAs (miRNAs) in endothelial cells (ECs).

Methods

These compounds were obtained through coating of CNTs with two polyamine polymers (i.e., polyethyleneimine, PEI and polyamidoamine dendrimer, PAMAM), followed by complexation with miRNA mimics. In particular, both PEI-coated and PAMAM-coated CNTs were able to stabilize miR-503 oligonucleotides and allowed an efficient delivery of polyplexes to ECs.

We further investigated the cellular internalization mechanism of polymer-coated CNTs compared to polymers alone. Using inhibitors of clathrin-mediated endocytosis (i.e, chlorpromazine) and caveolae-mediated uptake (i.e., filipin/genistein) we studied the internalization mechanisms of polymer-coated CNTs by different cell lines.

Results

FACS analysis suggested that these compounds may enter cells following different internalization pathways. In this contribution we will show the obtained preliminary results.

Discussion

Inhibition assays may contribute to understand the internalization processes by polyamines/dendrimers and polyamine-coated CNTs.

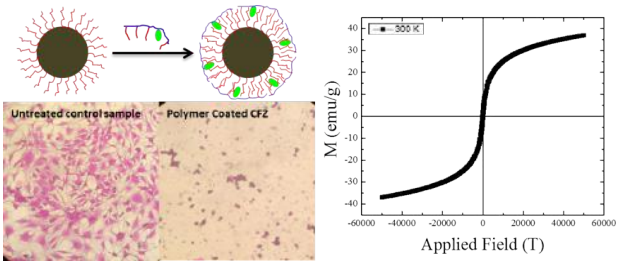
Synthesis of polymer coated Co0.5Zn0.5Fe2O4 nanoparticles and their cytotoxicity on human carcinoma cells

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 395

Dr. Zulqurnain Ali [1], ***Dr. Rashda Abbasi*** [2], ***Dr. Muhammad Atif*** [1], ***Ms. Javeria Arshad*** [1], ***Mr. Abdul Jabbar Khan*** [1], ***Dr. Nafees Ahmad*** [2]

1. *Air University,* ***2.*** *Institute of Biomedical and Genetic Engineering, Islamabad*

Over recent decades, the application of magnetic nanoparticles has been broadly investigated with their advance applications in the areas of biotechnology, medicinal drug delivery, magnetic fluid hyperthermia, and magnetic resonance imaging. Spinel ferrite nanoparticles are of importance because of their well-known unique optical, electronic and magnetic properties. Spinel ferrite nanoparticles have no preferred direction of magnetization, good saturation magnetization and high permeability. Being magnetically soft; they can easily magnetized and demagnetized, and electrically insulating. For this reason, in this study, cobalt zinc ferrite (Co0.5 Zn0.5 Fe2O4) magnetic nanoparticles (CZF) were used for further investigation. However, there are no data about biological activity of CZF, including their cellular uptake, toxicity, effects on cell proliferation, phenotype and functional activity. Therefore, these aspects remain a subject of particular interest. Multifunctional CZF were synthesized by sol gel method with high colloidal stability having room temperature ferromagnetism. The synthesis method is cost-effective, easy to scale up and reproducible. For biological applications CZF were polymer coated with amphiphilic polymer. The amphiphilic polymer can be functionalized with organic, organo-metallic or anticancer drug molecules; for this linkage the molecules need to bear a free amino group. Via the amino group these molecules are then directly attached to the maleic anhydride rings of the amphiphilic polymer. In this study 1% anhydride rings of amphiphilic polymer were reacted with fluorescent dye (cresyl violet perchlorate) used as model system for anti-cancer drug loaded polymer shell. The amount of functional molecule can be varied up to 25% of the anhydride rings, which provides greater affinity of drug loading in polymer shell. CFZ were further characterized by XRD, FTIR, UV-Vis spectroscopy and gel electrophoresis. The in vitro cytotoxicity of CZF was examined via the MTT assay on HepG2 (Liver Cancer cells). Concisely, 104 cells per well were seeded in to 96-well plates and exposed to different concentrations of CFZ for 24 h. The CFZ induce significant death of the carcinoma cells (HepG2). Stimulatingly, this seems to be a noteworthy improvement towards the ability of surface functionalized multifunctional CZF as carriers for drugs for anti-cancer therapy.



Ali abstract.png

Ratiometric real-time measurement of protein kinase activity with fluorophore labeled polyion complexes

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 402

Mr. Takanobu Nobori [1], ***Prof. Akihiro Kishimura*** [1], ***Prof. Takeshi Mori*** [1], ***Prof. Yoshiki Katayama*** [1]

1. *Department of Applied Chemistry, Kyushu University*

□Introduction□

The dysfunction of protein kinase (PK) activity results in various human diseases. Thus, a measurement method for PKs is not only valuable for understanding the molecular mechanism of complicated cellular events but also for diagnosis and drug discovery. Recently, fluorescence probes which get turn on during PK-mediated phosphorylation of substrate peptides are one of the attractive methods due to sharp contrast in simple optical readout systems. However, it is still difficult to realize quantitative real-time analysis and monitoring of PK activity in living cells. Therefore, we demonstrate a novel quantitative real-time detection system of PK activity. Our system is based on a polyion complex (PIC) nanoparticle embedded with a PK-responsive fluorescence signal and a second constant internal standard signal.

□Methods□

5-(and 6) -carboxytetramethyl-rhodamine (TAMRA) was modified to poly-L-aspartic acid (pAsp-TAMRA) as anionic polymer. As for cationic polymer, Cy5 and protein kinase Cα (PKCα) substrate peptide were modified to Dextran (Dex-Cy5-pep). After PICs formation of these polymers through electrostatic interaction, various concentrations of the PKCα were added and changes in the TAMRA and Cy5 fluorescence intensities were monitored.

□Results and Discussion□

Once pAsp-TAMRA and Dex-Cy5-pep formed the PICs, TAMRA fluorescence was selectively quenched and Cy5 fluorescence tended to be constant. This uneven quenching behavior of the two signals was controlled by adjusting each fluorophore content in each polymer. High content of TAMRA in pAsp-TAMRA (2.1 mol%) induced concentration quenching of TAMRA, while low content of Cy5 in Dex-Cy5-pep (0.4 mol%) did not show the quenching. In the monitoring of each fluorescence intensities with phosphorylation reaction of PICs, the recovery of TAMRA fluorescence intensity was observed with increasing reaction time and PKCα concentration. On the other hand, Cy5 fluorescence intensity was still constant. This result indicated that the electrostatic interaction between two types of polymers were weaken by phosphorylation and PICs were dissociated, which resulted in TAMRA fluorescence recovery from quenching. In this research, we realized this probe enables quantitative and real-time detection of PKCα activity. In near future, we will apply this PIC type probe to the measurement of PK activity in living cells.

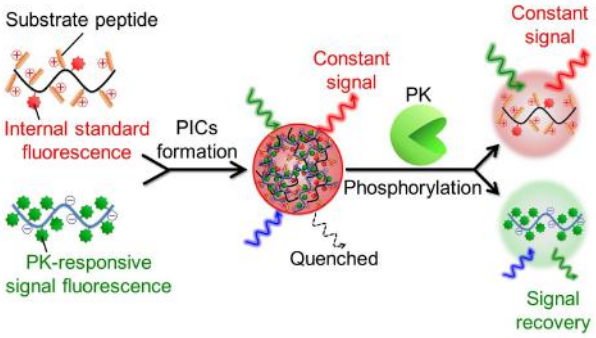


Figure for abstract.jpg

Synthetic hydrogels based on hybrid physically-chemically cross-linked networks

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 422

Dr. Maxime Grillaud [1], ***Mr. Maarten Bakker*** [1], ***Dr. Patricia Dankers*** [1]

1. *Eindhoven University of Technology*

Many polymers with different types of cross-linking have been designed to develop various hydrogels for industrial and fundamental applications. In particular, they have attracted significant interest in the field of regenerative medicine, as drug delivery vehicles or scaffolds for tissue engineering. However, the main challenging limitation for their extensive use is their low mechanical stability. Many recent efforts have been made to overcome this burden, in particular by the elaboration of double network (DN) hydrogels. Despite the promising results of the few existing physically-chemically cross-linked DN hydrogels, the preparation procedures require long photopolymerization time and toxic precursor materials. Moreover, only the comparison of the mechanical properties between the hybrid DN hydrogel made in very specific conditions with the corresponding two separate networks is studied. The fundamental understanding of the combination of two different cross-links for a rational design of new hybrid DN hydrogels is a challenge.

Herein, we provide synthetic strategies to tailor in-situ formation of DN hydrogels based on physically-chemically cross-linked networks. Three approaches to combine catalyst-free click reaction and hydrogen bonding for in-situ hydrogel formation will be described (see Figure). More in detail, the covalent cross-linking consists of the click reaction between azides and cyclooctyne derivatives, referred to as strain-promoted azide-alkyne coupling (SPAAC), and the non-covalent cross-linking is based on supramolecular interactions via a four-fold hydrogen bonding motif, the ureido-pyrimidinone (UPy) unit.

The mechanical properties of these new hydrogels are studied by rheology and their morphology are analyzed by confocal microscopy. A detailed comparison of the hydrogel properties is discussed according to the approach selected and the ratio of the two types of cross-linking. In particular, this work shows the benefits to associate the dynamicity afforded by supramolecular interactions for gel recovering with the strength provided by the covalent cross-links for enhanced material stiffness. We propose that this work offers both a novel versatile platform for the easy preparation of DN hydrogels under physiological conditions with tunable properties, and one more step in the fundamental understanding of structure-property relationship of physically-chemically cross-linked hydrogels.

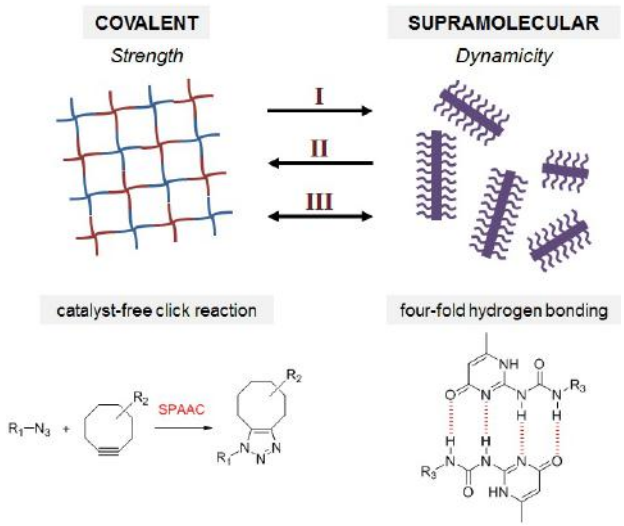


Figure abstract.jpg

A Novel niosome gene delivery approach for central nervous system disorders

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 438

Mr. Mohamed Mashal [1]***, Dr. Noha Attia*** [2]***, Prof. Gustavo Puras*** [3]***, Prof. Jon Zarate*** [4]***, Dr. Cristina Soto Sanchez*** [5]***, Prof. Eduardo Fernandez*** [5]***, Prof. Jose Luis Pedraz*** [3]

1. *University of the Basque Country, UPV/EHU, Spain,* ***2.*** *University of the Basque Country, UPV/EHU. University of Alexandria,* ***3.*** *University of the Basque Country, UPV/EHU,* ***4.*** *University of the Basque Country UPV/EHU,* ***5.*** *University of Miguel Hernandez*

Introduction

Glial cells are part of the nervous tissue where they nourish protect and regulate neuronal function. Therefore, they are ideal vehicle for gene therapy of brain diseases. Niosomes represent a recent safe approach for gene delivery purposes. Thus, our aim was to formulate an effective niosome formulation based on the cationic lipid N-[1-(2,3-dioleoyloxy)propyl]-N,N,N-trimethylammonium chloride (DOTMA) to deliver genes to nervous tissue.

Methods

Modified reverse phase evaporation technique was applied to obtain our niosomes. After their physicochemical characterization, niosomes were complexed with pCMS-EGFP reporter plasmid to form nioplexes. Afterwards, nioplexes were used to transfect NT2 cells as well as primary neuronal cells from rat embryonic cortex in vitro followed by studying their uptake and internalization pathways. Finally, Nioplexes were injected to transfect rat cerebral cortex in vivo.

Results

Nioplexes depicted appealing physicochemical properties as gene delivery vehicles. Additionally, they could transfect both NT2 and primary glial cells without compromising cell viability as detected by microscopy and flowcytometry. NT2 showed high uptake of nioplexes (59.6±7.01%), possibly due to their endocytosis by all the tested mechanisms; clathrin-mediated endocytosis (CME), caveolae-mediated endocytosis (CvME) and macropinocytosis. Nevertheless, their co-localization with both CME and lysosomes could explain the relatively lower transfection (up to 16%). When nioplexes were injected into rat cerebral cortex, they had tropism to transfect glial cells.

Conclusion

Thanks to its effectiveness to transfect cortical glial cells in rat brain without the known risks of viral vectors, our niosome vector could be an interesting novel non-viral formulation for gene therapy targeting severe diseases of the brain.

Proteins adsorption upon nanoparticles: from the physicochemical basis to the functional impacts

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 474

Mr. Laurent Marichal [1], ***Dr. Jean-philippe Renault*** [1], ***Dr. Jean-christophe Aude*** [2], ***Dr. Yves Boulard*** [3], ***Dr. Serge Pin*** [1], ***Dr. Jean Labarre*** [2]

1. *CEA Saclay, NIMBE,* ***2.*** *CEA Saclay, iBiTec-S,* ***3.*** *CEA Saclays, iBiTec-S*

The production and utilization of nanoparticles has been strongly increasing over the last decades, raising concerns regarding their biological effects and health hazards. Due to their nanometric size, nanoparticles have two major properties, (i) a high specific surface and (ii) the ability to cross biological barriers. Thus, they can interact with a large amount of biological compounds (especially proteins) and impact biological processes.

We study adsorption of proteins on silica nanoparticles (SiNP). We used yeast protein extracts in order to assess which kinds of proteins tend to adsorb on SiNP and which do not. Using proteomics (mass Spectrometry Shotgun analysis), we identified several hundreds of adsorbed and non-adsorbed proteins. The statistical comparative analysis of these two sets of proteins revealed the physicochemical determinants relevant for adsorption: the overrepresentation of arginine residues and of large disordered regions, as well as an impoverishment in secondary structures and hydrophobic aminoacids. These results are consistent with the notion that disorder and flexibility favor protein adsorption.

Using GO term analysis, we also evidenced that RNA binding proteins have a marked affinity for SiNP. We selected one of them, NPL3, a protein carrying poly(A) mRNA from nucleus to cytoplasm. We constructed two truncated mutants fused to GFP (see figure) and found that the C-ter region of NPL3 containing several repetitions of the Arg-Gly-Gly (RGG) motif is responsible for the high affinity of the protein for SiNP.

In vivo, the Arg residue of RGG motif is frequently dimethylated. We synthesized peptides (25-mer) containing RGG repetitions and some versions where Arg residues were dimethylated. The data indicate that Arg dimethylation significantly increases the affinity of the peptides for SiNP. We are currently planning to use calorimetry with these peptides in order to assess if the adsorption process, in these cases, is entropy or enthalpy driven.

Our data opens the way for predicting potential biological impacts of nanoparticles.

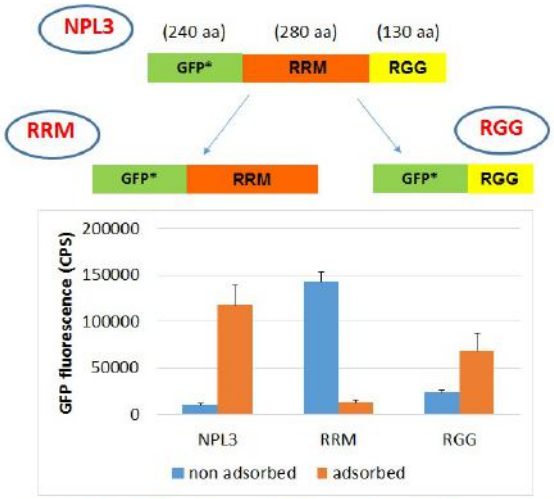


Figure: Representation of the adsorbed and non adsorbed fractions of the fluorescent proteins, namely NPL3 and 2 truncated mutants called RRM (RNA Recognition Motif) and RGG on silica nanoparticle. Total protein extracts were produced from yeast strains expressing the different fusion proteins.

Npl3 mutants affinity towards sinp.jpg

Flexible cortical Multi-electrode array implant for neural recording in minipig

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 475

Mr. Jean-Marie Mayaudon [1], ***Dr. Lionel Rousseau*** [2], ***Dr. Gaëlle Offranc Piret*** [1], ***Dr. Blaise Yvert*** [3]

1. *INSERM, BrainTech Laboratory U1205, Grenoble,* ***2.*** *ESIEE Paris, ESYCOM Université Paris-Est,* ***3.*** *Univ. Grenoble Alpes, BrainTech Laboratory U1205, Grenoble*

Multi-Electrode Arrays are key neuronal interfacing systems in neurophysiological and clinical research to better understand healthy and pathological neural network. In neuronal implant domain, one of the major challenge is to propose a biocompatible device that remain stable over a long period of time. Flexible implants are promising to reduce tissue damage and inflammation on the long term, as they limit the frictions at the implant tissue interface.

Based on the micro and nano technologies, we fabricate a fully flexible 256 microelectrodes. Several biocompatible polymers can be used to achieve the implant that allow to vary and test different implant thicknesses (range between 5 to 20 µm). The MEA implant is arranged in several independent strips that cover most of the cortical surface, and shape the brain regions in order to keep a close contact distance from neural tissue, reduce micromotions, and avoid implant displacement.

Electrochemical characterization of electrodes will be achieved and biocompatibility of the implant will be assessed on rat primary cortical cells cultures.

Performance and stability of this device will be tested for in-vivo cortical recording in the minipig.

Exudate Triggered Metal Corrosion for Self-Powered Wound Healing Application

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 512

Mr. Sun Woong Han [1], ***Mr. Keun Ho Lee*** [1], ***Mr. Tae Hoon Ki*** [1], ***Prof. Hong Koo Baik*** [1]

1. *Yonsei University*

We developed a galvanic wound healing patch that can promote skin wound healing without the use of any external electric devices. Electrical fields have been proven to modulate skin cell behavior to enhance skin wound healing. Our galvanic wound healing patch generated electrical fields by galvanic corrosion triggered electric stimulation for skin wound healing. The galvanic corrosion properties have been proven to modulate skin cell behavior to enhance skin wound healing. In vivo data demonstrated that the galvanic patch promoted the wound healing process through enhanced cellular metabolism and migration. This wound healing property may lead to a clinically relevant galvanic wound healing patch therapy for self-powered wound healing application.

Collagen glycation versus chronologically-aged fibroblasts in keratinocyte differentiation of reconstructed human skin

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 556

Ms. Roberta Rigon [1], ***Mr. Christian Hausmann*** [2], ***Mr. Christopher Wolff*** [2], ***Dr. Julia Tigges*** [3], ***Prof. Ellen Fritsche*** [3], ***Dr. Marlus Chorilli*** [4], ***Dr. Christian Zoschke*** [2], ***Prof. Monika Schäfer-korting*** [2]

1. *Universidade Estadual Paulista "Júlio de Mesquita Filho", Araraquara, Brazil and Institute of Pharmacy, Freie Universität Berlin, Berlin, Germany,* ***2.*** *Institute of Pharmacy, Freie Universität Berlin, Berlin, Germany,* ***3.*** *IUF - Leibniz Research Institute for Environmental Medicine, Düsseldorf, Germany,* ***4.*** *Department of Drugs and Medicines - Univ. Estadual Paulista Júlio de Mesquita Filho - UNESP - Araraquara/SP*

Skin morphology markedly changes over the lifespan, but the effects on the skin barrier function remain to be elucidated for nanocarrier-enhanced and –targeted topical drug delivery [1]. Herein, we investigated the effects of collagen glycation and fibroblast age, respectively, on keratinocyte differentiation in reconstructed human skin (RHS).

For glycation, collagen was incubated with 10 mM D-ribose for 3 weeks, dialyzed in water for 48 h, and analyzed by fluorescence spectroscopy [2]. Glycated collagen was mixed with non-glycated collagen 1:1 and used for RHS. Juvenile keratinocytes were used for the epidermal compartment of all RHS. Fibroblasts for glycated and juvenile (non-glycated) RHS were from juvenile foreskin; too, in addition we used fibroblasts from mammary reduction surgeries of 60- to 70-year-old female donors. RHS were cultured for two weeks after the exposure to the air-liquid-interface and then subjected to reflectance confocal microscopy, immunofluorescence staining or skin surface pH analysis.

We detected 30% of collagen glycation by fluorescence spectroscopy and significant amounts of carboxymethyl lysine staining in glycated models. Stratum corneum thickness increased in glycated and aged RHS 2-fold compared to juvenile RHS with the most dense stratum corneum appearance in glycated RHS. The surface pH of glycated models exceeded the surface pH of RHS with chronologically-aged fibroblasts and juvenile RHS. Expression of keratinocyte terminal differentiation markers like filaggrin and loricrin as well as integrin expression was enhanced in glycated RHS whilst dermal thickness decreased in glycated models compared to juvenile and aged RHS (Figure 1), which is in accordance to literature [3].

Taken together, both approaches mimic hallmarks of cutaneous aging. Collagen glycation affects keratinocyte differentiation more extensively than the replacement of juvenile by chronologically-aged fibroblasts, highlighting the impact of glycation for the skin barrier function. Future studies will evaluate the functionality of the skin barrier.

References

[1] Biniek et al. (2015). J Dermatol Sci 80: 94-101.

[2] Pennacchi et al. (2015). Tissue Eng Part A 21:2417-25.

[3] Pageon et al. (2015). BioResearch Open Access, 4.1.

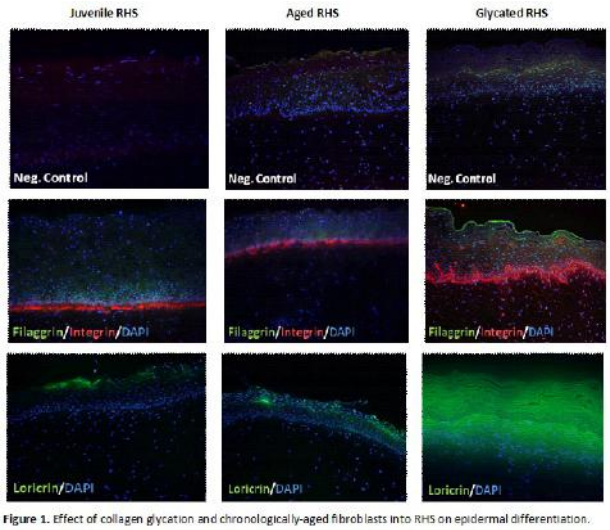


Figure 1. Effect of collagen glycation and chronologically-aged fibroblasts into RHS on epidermal differentiation.

Figure 1. effect of collagen glycation and chronologically-aged fibroblasts into rhs on epidermal differentiation..jpg

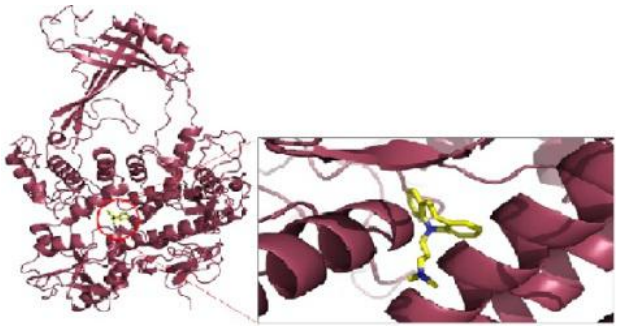
Nanoformulation of imipramine loaded resealed erythrocytes as potent anti-leishmanial, which targets unique `prokaryotic TopA' homolog in Leishmania

Thursday, 29th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 97

Ms. Sumedha Mukherjee [1], ***Ms. Devyani Shukla*** [1], ***Dr. Somdeb Bosedasgupta*** [1]

1. *IIT BHU, Varanasi*

Introduction:
Uncontrolled use of traditional anti-leishmanials leading to the emergence of drug resistant parasites has necessitated the search for potent drugs against novel drug targets clear and evident. A serendipitous BLAST search using prokaryotic TopA sequence led us to identify its homolog, LdBPK_210180.1 in Leishmania. The sequence corresponds to a unique Topoisomerase IA (LdTopIA), having prokaryotic homologs and being absent in higher eukaryotes. Previously anti-depressants have been shown to exhibit anti-leishmanial activity and imipramine, which is one such anti-depressant has been reported to inhibit mycobacterial Topoisomerase I (MttopoI), so we thought of testing efficacy of imipramine as an inhibitory agent against LdTopIA.
Methods:
Sequence similarity of E.coli TopA and LdTopIA was established using CLUSTAL-W and homology modeling of LdTopIA was performed using SWISS-MODEL using E.coli TopA as template (PDB ID-1CY1). Functional complementation studies were performed in RFM 475 (an E.coli TopoI null-DNA gyrasets mutant strain) in presence and absence of imipramine. In-silico docking was also studied for imipramine and LdTopIA. Nanoformulation of imipramine was carried out for entrapment in resealed erythrocytes for efficient delivery to the effected organs.
Results:
Homology modeling and functional characterization shows LdTopIA has active site residues similar to its prokaryotic homologs. Though the enzyme complements E.coli TopA in RFM 475 strain, it fails to do the same in presence of imipramine. In-silico docking studies also show that imipramine binds to the active site of LdTopIA suggesting that imipramine is a potent inhibitor of LdTopIA.
Discussion:
The results establish LdTopIA as a unique drug target capable of killing the parasite while respite the host. Since, liver and spleen are the effected organs in visceral leishmaniasis, nanoformulation of imipramine-loaded resealed erythrocytes can be an effective therapy to combat the disease as the reticuloendothelial system readily phagocytoses such RBCs. Such strategy can lead to personalized medicines where one's own red blood cells can be used as drug delivery vessel and thereby prove to be a successful new strategy for treating leishmaniasis.



Imipramine binds to ldtopia active site.jpg

Platinum nanoparticles as multifunctional active nanocarriers integrating the function of high-performance antioxidant drugs

Thursday, 29th September - 16:00 - Targeted drug delivery and Nanocarriers - Nanomedecine for cancer diagnosis & therapy - Amphitheatre 25 - Oral presentation - Abstract ID: 252

Ms. Deborah Pedone [1], ***Dr. Elisa De Luca*** [2], ***Dr. Mauro Moglianetti*** [2], ***Dr. Roberto Marotta*** [3], ***Mr. Tiziano Catelani*** [3], ***Dr. Barbara Sartori*** [4], ***Dr. Heinz Amenitsch*** [4], ***Prof. Saverio Francesco Retta*** [5], ***Dr. Pier Paolo Pompa*** [6]

1. *Istituto Italiano di Tecnologia, Center for Biomolecular Nanotechnologies; University of Salento, Department of Engineering for Innovation,* ***2.*** *Istituto Italiano di Tecnologia, Center for Biomolecular Nanotechnologies,* ***3.*** *Electron Microscopy Laboratory, Nanochemistry Department, Istituto Italiano di Tecnologia,* ***4.*** *Graz University of Technology, Institute of Inorganic Chemistry,* ***5.*** *Department of Clinical and Biological Sciences, University of Torino,* ***6.*** *Istituto Italiano di Tecnologia*

Introduction

In recent years, nanomaterials that mimic natural enzymes (nanozymes) have elicited huge interest in nanomedicine (Wei and Wang, Chemical Society Reviews, 2013). In this work, we explored the use of citrate-capped platinum nanoparticles (Pt NPs) as nanozymes that act simultaneously as drugs and carriers in oxidative stress-mediated diseases.

Methods

We synthesized citrate-capped Pt NPs of 5 and 20 nm of diameter and performed a systematic characterization of their superoxide dismutase (SOD)-, catalase (CAT)- and peroxidase (HRP)- like activities in cell-free environment.

As the combined function of drug and carrier requires that Pt NPs are cytocompatibles, we performed a systematic toxicity assessment (WST-1, LDH leakage, TUNEL, and DCF assay), investigating the internalization of Pt NPs and their subcellular localization by TEM and ICP-AES analyses.

Finally, to test the hypothesis that citrate-capped Pt NPs can act as a scavenging material in biological systems, we tested Pt NPs on the cellular model of a cerebrovascular disease, namely Cerebral Cavernous Malformation (CCM), characterized by an abnormal angiogenesis and associated with a significant increase in intracellular reactive oxygen species (ROS) levels.

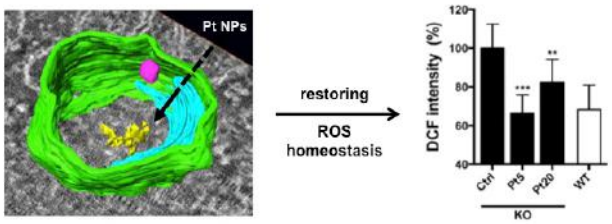
Results and Discussion

We showed that pure, monodisperse, and endotoxin-free Pt NPs do not exert any toxicity on several cell lines, independently of the size. We proved that the absence of toxicity of Pt NPs is related to their compartmentalization and stability within the endo/lysosomal vesicles and to the absence of release of Pt ions.

We demonstrated, for the first time, that Pt nanozymes are capable to restore physiological ROS homeostasis in a real experimental model of CCM disease, founding that Pt NPs can completely recover the cellular phenotype, similar to that of wild type cells (Fig.1).

This is possible because of the strong and broad antioxidant nanozyme activity of Pt NPs, which are simultaneously endowed with strong CAT-, HRP-, and SOD-like activities, with superior performance than natural enzymes (Moglianetti et al., Nanoscale, 2016).

These findings are important and of broad interest, and open up novel perspectives in nanomedicine for the development of multifunctional active nanocarriers integrating the function of high-performance antioxidant drugs with strong potential for therapies of complex oxidative stress-related diseases.



Pt nps restore ros homeostasis in cellular model of oxidative stress mediated diseases .jpg

Transport of Liposome Encapsulated Drugs in Voxelized Computational Model of Brain Tumors

Thursday, 29th September - 16:25 - Targeted drug delivery and Nanocarriers - Nanomedecine for cancer diagnosis & therapy - Amphitheatre 25 - Oral presentation - Abstract ID: 239

Mr. Ajay Bhandari [1], ***Dr. Ankit Bansal*** [2], ***Dr. Anup Singh*** [3], ***Dr. Niraj Sinha*** [1]

1. *Indian Institute of Technology, Kanpur,* ***2.*** *Indian Institute of Technology, Roorkee,* ***3.*** *Indian Institute of Technology, Delhi*

Introduction:

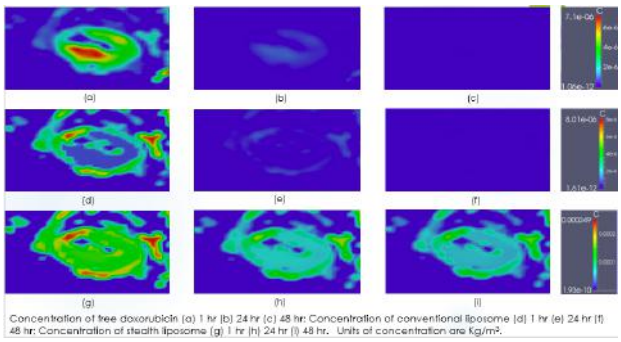
Cancer is one of the leading causes of death all over the world. Various treatment strategies are now a days used for treatment of cancers such as chemotherapy, radiotherapy and surgical removal. Liposome mediated drug delivery has emerged as an excellent anticancer therapy due to its ability to deliver drugs at site of action and reducing the chances of side effects to the healthy tissues. Computational modelling provides us a non-invasive approach to predict the transport mechanism of liposomes to tumor site and their accumulation within tumor tissues. Aim of this study is to perform computer simulations to qualitatively and quantitatively analyze the deposition of liposomes in human brain tumors. Additionally, the size effect is incorporated by considering stealth (PEG encapsulated doxorubicin) and conventional liposomes. Their accumulation results are compared with the free drug (doxorubicin).

Methods:

In this study computational fluid dynamics has been used to develop a voxelized model based on DCE-MRI. Complete analogy is maintained between MRI slice and CFD slice by assigning all the variables voxelwise. All the fluid flow and solute transport equations have been simulated to predict the transport of liposomes to tumor site. To the best of our knowledge this is the first study to predict the transport of liposomes in realistic heterogeneous vasculature of tumors computationally.

Results and Discussions:

Simulation has been performed for 48 hours. Results indicate that stealth liposomes accumulate more and remain for longer period of time in tumors as compared to conventional liposomes and free drug. Stealth liposome concentration has been estimated approximately 20 fold more than that of free drug and it remains in the tumor tissue for longer period as compared to conventional liposomes and free drug. These values have been validated with those in literature. Reason behind stealth liposome more accumulation in tumors is their slower plasma and higher vascular clearance. This increases the therapeutic effect of drug and also their probability of killing tumor cells due to long drug exposure. This computational study potentially enables medical professionals to forecast the overall effectiveness of the drug and optimize treatment strategy for each patient.



Contour plots of concentration.png

Sensitization of sarcoma tumors with short chain sphingolipid liposomes

Thursday, 29th September - 16:50 - Targeted drug delivery and Nanocarriers - Nanomedecine for cancer diagnosis & therapy - Amphitheatre 25 - Oral presentation - Abstract ID: 510

Dr. Sara Zalba Oteiza [1], ***Mr. Joost A. P. Rens*** [1], ***Dr. Jeroen Rovers*** [2], ***Prof. Marcel Verheij*** [3], ***Dr. Timo L. M. Ten Hagen*** [1]

1. *Erasmus Medical Center,* ***2.*** *Ceronco Biosciences,* ***3.*** *Antoni van Leeuwenhoek*

Introduction: Multidrug resistance (MDR) is one of the main challenges in cancer chemotherapy. The over-expression of multidrug transporters like P glycoprotein (Pgp) is a common cause of resistance1. Liposomes enriched in short chain sphingolipids (SCS) may interact with cancer cell membranes improving permeability to encapsulated drugs2-4. Our aim is to evaluate the effect of SCS liposomes in resistant tumor cells with the aim of improve their chemo-sensitivity.

Methods: MES-SA/MX2 is a resistant human utero sarcoma cell line derived from MES-SA, with Pgp over-expression. Liposomes loaded with doxorubicin were enriched (SCS-LP) or not (LP) with SCS, characterized and incubated with both cell lines to evaluate drug internalization and cytotoxic effect. Biodistribution, pharmacokinetics and efficacy was evaluated in xenograft tumor bearing mice with the above mentioned sensitive and resistant cell lines.

Results: In vitro assays showed a higher effect of SCS-LP in MES-SA/MX2, which was related with an increase in the sensitivity to doxorubicin. Actually SCS-LP were able to revert their resistance compared to sensitive cell results. On the MES-SA cell line all liposomes had a similar effect under all conditions tested. A higher accumulation of doxorubicin in resistant tumors after SCS-LP administration was observed in mice bearing these tumors subcutaneously, compared to the sensitive tumor and LP. Besides, nuclear accumulation of doxorubicin was superior in resistant tumors under SCS-LP treatment. Finally, enriched liposomes showed a faster clearance but were able to control the tumor growth more efficiently than non SCS liposomes in resistant tumors, whereas in sensitive tumor no difference was found.

Discussion: Based on our in vitro and in vivo results we conclude that the use of doxorubicin-SCS-LP reverts the resistance of MES-SA/MX2. Our data indicates that sensitivity to the drug is improved through bypassing or compensation of Pgp activity. Although the formulation needs to be optimized, e.g. to avoid serum instability, these liposomes represent a very promising tool to treat cancer, especially MDR tumors.

1.Adv Drug Deliv Rev. 2013 Nov;65(13-14):1852-65.

2. J Biomed Nanotechnol. 2016 Apr;12(4):630-44.

3. Pharm Res. 2015 Apr;32(4):1354-67.

4. Pharm Res. 2013 Jul;30(7):1883-95.

Immune checkpoint blockade in melanoma by new targeted Doxorubicin immunoliposomes

Thursday, 29th September - 17:15 - Targeted drug delivery and Nanocarriers - Nanomedecine for cancer diagnosis & therapy - Amphitheatre 25 - Oral presentation - Abstract ID: 511

Mrs. María Merino Díaz [1], ***Mrs. Ana Margarita Contreras Sandoval*** [1], ***Dr. Noelia Casares*** [2], ***Dr. Iñaki Troconiz*** [1], ***Dr. Pedro Berraondo*** [2], ***Dr. Timo L. M. Ten Hagen*** [3], ***Dr. Sara Zalba Oteiza*** [3], ***Dr. Maria J Garrido*** [1]

1. *University of Navarra,* ***2.*** *CIMA,* ***3.*** *Erasmus Medical Center*

Introduction: Programmed Cell Death Ligand 1(PD-L1), expressed in most solid tumors, binds to T-cells through its receptor PD-1, leading to the immunoresistance development and tumor progression [1]. Anti-PD-L1 monoclonal antibody (mAb) blocks this ligand promoting antitumor activity of T-cells [2]. Moreover, Doxorubicin (Dox) liposomes improve the therapeutic effect over free drug. Both therapies can be combined in a single formulation of Dox liposomes decorated with anti-PD-L1 Fab' fragments. Thus, the aim of this work is to develop and characterize Dox targeted liposomes and evaluate its antitumor effect in vitro and in vivo.

Methods: Dox liposomes (LPDOX) were prepared by film hydration method [3] and drug loaded by sulphate gradient [4]. DSPE-PEG2000-Mal micelles were coupled to anti-PD-L1-Fab' fragments and incorporated in pre-formed LPDOX with post insertion method, obtaining targeted liposomes (LPDOXFab'). After physicochemical characterization, drug release was measured in FBS at 37 ºC for 1h. B16 OVA melanoma mouse cell line was used for in vitro/in vivo studies. In vitro cytotoxicity (IC50) was obtained after 24h of drug exposure and Dox cell uptake was measured by FACS and confocal microscopy after 4h of treatment. Finally, in vivo antitumor efficacy was studied in tumour bearing mice.

Results: LPDPXFab' and LPDOX were around 130 nm, PDI < 0.1 and 95±7.5% of encapsulation. Ligand conjugation was 40±7.4%. Drug release reached 20% after 1h in FBS. Targeted liposomes were able to bind specifically PD-L1, showing no internalization. IC50 values for both formulations were similar after 24h of exposure. There was a delay in melanoma tumor growth with no significant differences in both liposomes. However, survival for LPDOXFab' was higher, suggesting a benefit that deserves to be explored.

Conclusion: LPDPXFab' have been successfully developed using the post-insertion method. In vitro and in vivo studies revealed similar efficacy for both formulations. However, the difference in the survival suggests an improvement of the targeted formulation that will be deeper examined in further studies.

[1] Crit Rev Oncol Hematol. 2014 Jan; 89(1):140-65.

[2] Eur J Cancer. 2013 Sep; 49(14):2968-71.

[3] Eur J Pharm Biopharm. 2012 Jun; 81(2):273-80.

[4] J Control Release. 2012 Jun 10; 160(2): 117-34.

Titanate nanotubes as new preclinical theranostic platform against prostate cancer: vectorization and immobilization of docetaxel or of gold nanoparticles

Thursday, 29th September - 17:40 - Targeted drug delivery and Nanocarriers - Nanomedecine for cancer diagnosis & therapy - Amphitheatre 25 - Oral presentation - Abstract ID: 14

Mr. Alexis Loiseau [1], ***Dr. Julien Boudon*** [1], ***Dr. Céline Mirjolet*** [2], ***Prof. Gilles Créhange*** [2], ***Prof. Stéphane Roux*** [1], ***Prof. Nadine Millot*** [1]

1. *Université Bourgogne Franche-Comté,* ***2.*** *CGFL*

Titanate nanotubes (TiONts) are synthetized by a hydrothermal process [1]. Their uncommon morphology obtained by controlled parameters allows them to be internalized more easily into cells. This permits their use as novel transfection agents [2] without inducing cytotoxicity while providing a radiosensitization effect [3]. These TiONts are combined to docetaxel (DTX), a molecule widely used for the treatment of cancers. Currently, injected drugs only reach very weakly tumor sites. In the last decade, the development of nanotechnologies has offered a new strategy to vectorize an active substance. In this work, two developments of nanohybrids are presented to fight against prostate cancer [4]: the first approach consists in combining TiONts and DTX and in a second one TiONts and gold nanoparticles (AuNPs) are combined together. This project is based on intraprostatic injection of the nanohybrids.

A particular attention was paid on the elaboration of functionalized-TiONts nanohybrids. On the one hand, docetaxel molecules were grafted by an original pathway onto TiONts. DOTA macrocycles were also grafted on the latter, allowing a radiotracer to be chelated, in order to evaluate the biodistribution of TiONts in mice by nuclear imaging. Biodistribution kinetics showed that more than 70% of nanohybrids were localized into the tumor 96 hours after injection. Mice receiving nanohybrid-RT (Radiation Therapy) exhibited a significant tumor growth delay compared to mice receiving free DXL-RT [5]. On the other hand, AuNPs functionalized by DTDTPA molecules are grafted on TiONts. This combination confers not only the nanovectorization via TiONts but also therapeutic effect via AuNPs [6]. In both cases, the suspension stability of nanohybrids was increased by the addition of biocompatible polyethylene oxide polymers and physico-chemical characterizations were realized to assert each functionalization step of our engineered nanohybrids.

References:

[1] A.-L. Papa et al., J. Phys. Chem. C, 113 (2009)

[2] A.-L. Papa et al., Nanotoxicology, 7, 6 (2013)

[3] C. Mirjolet et al., Radiother. Oncol., 108, 1 (2013)

[4] J. Boudon et al., Nanomedecine. O. C. Press: chapt 16

[5] C. Mirjolet et al., Eur. J. Cancer, 50, 6 (2014)

[6] C. Alric et al., Gold Bulletin, 41, 2 (2008)

The endothelial glycocalyx controls interactions of nanoparticles with the endothelium and their translocation across the blood-tissue border

Thursday, 29th September - 16:00 - Targeted drug delivery and Nanocarriers - Tower 24 - Room 101 - Oral presentation - Abstract ID: 414

Dr. Bernd Uhl [1], ***Dr. Stephanie Hirn*** [1], ***Mr. Roland Immler*** [1], ***Mrs. Karina Mildner*** [2], ***Dr. Leonhard Möckl*** [3], ***Prof. Markus Sperandio*** [1], ***Prof. Christoph Bräuchle*** [3], ***Dr. Christoph Reichel*** [1], ***Dr. Dagmar Zeuschner*** [2], ***Prof. Fritz Krombach*** [1]

1. *Walter Brendel Centre of Experimental Medicine, Klinikum der Universität München, Ludwig-Maximilians-Universität München, Munich, Germany,* ***2.*** *Electron Microscopy Unit, Max Planck Institute for Molecular Biomedicine, Münster, Germany,* ***3.*** *Department of Chemistry and Center for NanoScience (CeNS), Ludwig-Maximilians-Universität München, Munich, Germany*

A prerequisite for the use of nanoparticles (NPs) in diagnostic and therapeutic applications is the targeted delivery to the site of injury. The endothelial glycocalyx (eGCX) is a glycoprotein-polysaccharide meshwork coating the luminal surface of the vasculature. Its role for the regulation of interactions between blood-borne NPs and the microvascular endothelium, however, remains obscure.

Multiphoton in vivo microscopy was employed on the cremasteric microvasculature of wildtype mice to analyze the structure of the eGCX and the deposition of NPs. The eGCX was visualized using labeled wheat germ agglutinin. Carboxyl quantum dots (QDs) were chosen as model NPs. Heparinase, hyaluronidase, and neuraminidase were used to systemically degrade the eGCX. Direct binding of NPs to endothelial molecules was examined in vitro using flow chambers. The uptake and translocation of NPs were addressed in vivo employing transmission electron microscopy (TEM). The effect of ischemia-reperfusion (I/R) injury on the eGCX and the deposition of NPs were examined using an I/R model.

In the microvasculature of control animals, a high eGCX layer was found in all vessel segments, while interactions of QDs with the endothelium were low. Upon enzymatic degradation of the eGCX, however, its size was significantly reduced and NP-endothelium interactions increased considerably. Moreover, endothelial uptake and transendothelial translocation of NPs after enzymatic degradation of the eGCX were demonstrated in vivo using TEM. Binding of QDs to endothelial signaling and adhesion molecules, which were exposed to the bloodstream upon enzymatic degradation of the eGCX, was observed by in vivo microscopy and confirmed in flow chambers. Employing I/R on the cremaster muscle caused a significant reduction of the eGCX and a dramatic increase in NP-endothelium interactions.

Consequently, the eGCX effectively controls NP-endothelium interactions in the microvasculature. It was found to physically cover endothelial adhesion and signaling molecules thereby preventing endothelial attachment, uptake, and translocation of NPs through different layers of the vessel wall. Conversely, degradation of the eGCX under pathologic conditions promoted interactions of NPs with microvascular endothelial cells thus identifying the injured eGCX as an essential element of the blood-tissue border facilitating the targeted delivery of nanomaterials to diseased tissue.

Magnetic hyperthermic response of nanocomposites based on PEG2000-gallol-coated iron oxide nanoparticles dispersed in poly(N-isopropylacrylamide-co-acrylamide) hydrogels

Thursday, 29th September - 16:25 - Targeted drug delivery and Nanocarriers - Tower 24 - Room 101 - Oral presentation - Abstract ID: 463

Mr. Kevin C. Behan [1], ***Ms. Sarah M. Martyn*** [1], ***Dr. Aylvin A. Dias*** [2], ***Prof. Andreas Heise*** [3], ***Dr. Dermot F. Brougham*** [4]

1. *Dublin City University, School of Chemical Sciences, Glasnevin, Dublin 9,* ***2.*** *DSM Ahead, Urmonderbaan 22, 6167 RD Sittard-Geleen,* ***3.*** *Royal College of Surgeons, Department of Pharmaceutical and Medicinal Chemistry, 123 St. Stephen's Green, Dublin 2,* ***4.*** *University College Dublin, School of Chemistry, Stillorgan Road, Belfield, Dublin 4*

Nanoparticles can be incorporated into hydrogel matrices by various means. Non-covalent methods such as blending and in situ co-precipitation are straightforward, but can result in uncontrolled diffusion of the nanoparticles out of the gel. Covalent methods graft reactive groups onto the nanoparticle surface, which can then act as nano-crosslinkers, forming stable networks. [1]

Superparamagnetic iron oxide nanoparticles were synthesised step-wise from 6 nm seeds, according to the method by Sun. The hydrophobic oleic acid and oleylamine ligands were exchanged with the cathecol-bearing methoxy-PEG2000-gallol. These modified iron oxide nanoparticles could be transferred almost quantitatively into ethanol.

Subsequently, the methoxy-PEG2000-gallol coated nanoparticles were mixed with a solution containing various amounts of N-isopropyacrylamide, acrylamide, PEG700-diacrylate, ammonium persulfate, and TEMED. The solutions underwent thermal radical polymerisation to obtain the magnetic hydrogel nanocomposite. After purification, the gels were exposed to an AC magnetic field (475 kHz, 12.44 mT). Deswelling with respect to nanoparticle loading and acrylamide concentration was investigated. The methoxy-PEG chains on the nanoparticle surface ensure facile dispersion, a stable nanocomposite, with reproducible heating profile and deswelling behaviour.

Dhyd of the iron oxide nanoparticles was 44 nm and DTEM was 12 nm. For gels containing 200 µL of nanoparticle suspension (22.6 mM Fe, Tf=24.3°C), the deswelling ratio in the AC field was 8.6, 7.3, and 5.1%, respective of increasing acrylamide content. For gels containing 400 µL of nanoparticle suspension (45.1 mM Fe, Tf=32.1°C), the deswelling ratio was 39.7, 26.3, and 19.0%, respectively. For gels containing 800 µL of nanoparticle suspension (90.3 mM Fe, Tf=35.4°C), the deswelling ratio was 45.4, 43.1, and 26.4%, respectively.

The amount of acrylamide influences both the rates of swelling and deswelling in the hydrogel. Increasing iron oxide nanoparticle loading increases the temperature of the gels and accelerates the deswelling further. Both swelling and deswelling are reproducible for a given gel. It is planned to investigate deswelling of the gels at a baseline temperature of 37 °C.

[1] Li, Y., Huang, G., Zhang, X., Li, B., Chen, Y., Lu, T., Lu, T. J. and Xu, F. (2013), Magnetic Hydrogels and Their Potential Biomedical Applications. Adv. Funct. Mater., 23: 660–672. doi:10.1002/adfm.201201708

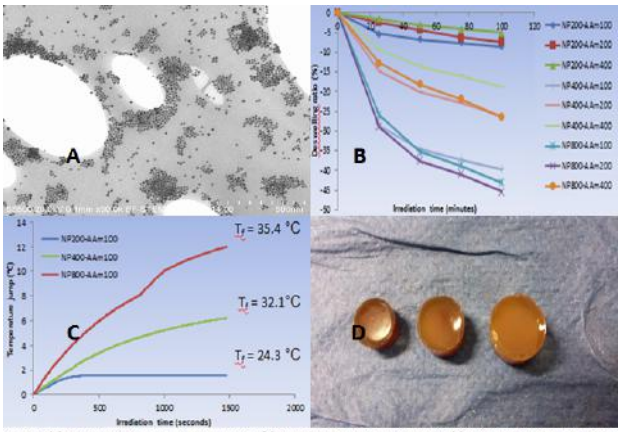


Figure 1: (A) TEM image of 12 nm iron oxide nanoparticles. (B) Deswelling ratios of magnetic hydrogels. (C) Heating profiles of gels as function of [Fe]. (D) Deswelling state based on acrylamide content (left: AAm100; middle: AAm200; right: AAm400)

Abstract fig.png

Increasing the stability and antimicrobial activity of antimicrobial peptides after association to Lipid nanocapsules

Thursday, 29th September - 16:50 - Targeted drug delivery and Nanocarriers - Tower 24 - Room 101 - Oral presentation - Abstract ID: 53

Ms. Nada Matougui [1], ***Dr. Anne Claire Groo*** [2], ***Dr. Anita Umerska*** [1], ***Dr. Helena Bysell*** [3], ***Prof. Patrick Saulnier*** [1]

1. *Université Angers, UMR-S1066 Angers, France,* ***2.*** *Normandie Univ, UNICAEN, Centre d'Etudes et de Recherche sur le Médicament de Normandie (CERMN), 14000 Caen, France,* ***3.*** *SP Technical Research Institute of Sweden*

Introduction:

Resistant infections are estimated to be responsible of 700.000 deaths worldwide. These figures, often underestimated highlight the importance of the antimicrobial resistance (AMR). Education toward the misuse of antibiotics is no more sufficient. Finding new therapies to overcome resistance becomes imperative. Antimicrobial peptides are under the spotlight. Their ability to act rapidly, at low concentrations and in a not specific way makes them interesting antibiotic molecules with less potential to induce resistance. Nevertheless, their development is hampered by physico-chemical and biological instabilities.

The aim of this work is to explore the potential of lipid nanocapsules (LNCs) to deliver AA230, DPK060 and LL37, three cationic, hydrophilic and biosourced antimicrobial peptides.

Methods:

Peptide adsorption on the surface of the LNCs (LNC-AMP) and/or encapsulation within their lipidic core (LNC-RM-AMP) was performed. The in vitro antimicrobial activity of the AMPs and nanoformulated AMPs was assessed by the determination of the minimal inhibitory concentrations against different Gram (-) and Gram (+) bacteria and The kinectics of the inhibitory effect was conducted for each formulation against the sensitive strains. Proteolysis assays was performed to invetigate peptides stability after association.

Results:

The results showed a good association efficiency after adsorption to the surface of LNCs. The encapsulation in the core succeeded except for the peptide AA230. The antimicrobial activity was preserved in all cases. Moreover, a potentiation of the in vitro anti-bacterial activity, reflected by a lower minimal inhibitory concentrations (MICs) was observed for the adsorbed peptides. The time kill-kinetics studies confirm the bactericidal killing of the nanoformulated peptides at concentrations up to 8 times lower than the MIC of the peptide alone. Moreover, proteolysis assays performed with trypsin as model enzyme, demonstrated a partial protection of adsorbed LL37 against proteolytic degradation compared to the plain peptide over 4hours.

Discussion:

Overall, these data indicated that LNCs are promising nanocarriers for the delivery of AMPs. The two strategies studied are to be investigated. The better knowledge of the peptide properties and the exploration of the mechanism of action responsible of the synergistic effect will definitely contribute to consolidate the potential of such nanoformulated peptides as next-generation antibiotics.

Bacterial strains	Minimum Inhibitory Concentrations (µg/mL)					
	LL37	LNC-LL37	DPK060	LNC-DPK060	AA230	LNC-AA230
Staphylococcus aureus	8-16	8-16	4	2-4		
Methicillin-resistant Staphylococcus aureus clinical strain (MRSA)	8-16	4	4	1		
Pseudomonas aeruginosa	8-16	16	8	8-16	2-4	1
Pseudomonas aeruginosa clinical strain	8-16	4	16	4-8	4-8	2
Escherichia coli ATCC 25922	16	8	8	4	2-4	1
Escherichia coli clinical strain	16	8	4-8	2-4	1-2	0.5
Acinetobacter baumannii ATCC BAA-1710	16	4	4-8	4-8	2-4	0.25

Minimal inhibitory concentrations of native and associated antimicrobial peptides.png

Biological Recognition of Biomolecular Corona

Thursday, 29th September - 17:15 - Targeted drug delivery and Nanocarriers - Tower 24 - Room 101 - Oral presentation - Abstract ID: 261

Ms. Sandra Lara [1], Ms. Fatima Alnasser [1], Dr. Ester Polo [1], Dr. David Garry [1], Ms. Maria-cristina Lo-giudice [1], Dr. Delyan Hristov [1], Dr. Yan Yan [1], Prof. Kenneth A. Dawson [1]

1. *Centre For BioNano Interactions (CBNI), School of Chemistry and Chemical Biology, University College Dublin*

Introduction

When the nanoparticle surface comes in contact with a biological milieu, adsorption processes may lead to a well-defined surface-assembly of biomolecules derived from the environment. Appropriately presented, and sufficiently long-lived ('hard corona') biomolecular motifs presented at the surface would define how a nanoparticle first interacts with and is recognised by cells. Besides their fundamental importance, such questions likely define many key impacts on efficacy of nanoparticle-based therapies, including their bio-distribution and nanomedicine targeting. For instance, well-defined active processes driven by receptors on the specialized cells of the liver could drive nanoparticle accumulation. However, little is known of the molecular organisation of various proteins in the corona. Hence, the detailed molecular interactions between the presented protein motifs and receptors are yet to be elucidated.

Methods

Here we present a systematic method to advance such hypotheses, by direct investigation of nanoparticle-receptor interactions in biological milieu. Firstly, we have built a cell receptor library by transfection receptor of interest to host cells. This system allows us to examine the dependence of nanoparticle uptake to a given receptor in appropriate biological milieu. In addition, this approach overcomes the limitations that some receptors cannot be isolated in a functionally relevant form, as their expression levels are interdependent in endogenously expressed cells. In parallel, we also have developed tools by using monoclonal antibodies to detect epitope presentation on the nanoparticle surface. Altogether, we have been able to characterise the biomolecular corona with molecular details and their interactions with different receptors.

Results and Discussion

In this presentation, we will illustrate the recognition of biomolecular corona by a couple of key receptors, low-density-lipoprotein receptor (LDLR) and Fc-gamma receptor I (FcγRI), which are abundant in the liver. By using both flow cytometry and microscopy, we have shown that the 'labelling' of nanoparticles by biomolecular adsorption processes allows for ubiquitous nanoparticle multi-pathway involvement in biological processes, which governs the liver accumulation of nanoparticles.

Synthesis-Dependent Surface Defects and Morphology of Hematite Nanoparticles and Their Effect on Cytotoxicity in Vitro

Thursday, 29th September - 17:40 - Targeted drug delivery and Nanocarriers - Tower 24 - Room 101 - Oral presentation - Abstract ID: 82

Mr. Dean Cardillo [1], ***Dr. Moeava Tehei*** [1], ***Dr. Md Shahriar Hossain*** [1], ***Mr. Md Monirul Islam*** [1], ***Ms. Kathrin Bogusz*** [1], ***Dr. Dongqi Shi*** [1], ***Dr. David Mitchell*** [1], ***Prof. Michael Lerch*** [1], ***Prof. Anatoly Rosenfeld*** [1], ***Dr. Stéphanie Corde*** [2], ***Dr. Konstantin Konstantinov*** [1]

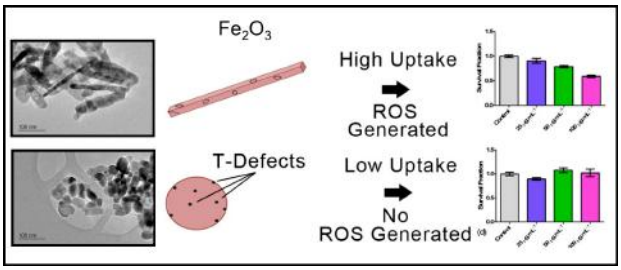
1. *University of Wollongong,* ***2.*** *Prince of Wales Hospital*

In this study we investigated the toxicity of hematite nanoparticles on the Madin-Darby Canine Kidney (MDCK) cell line. Hematite precursors were prepared using precipitation and spray precipitation, which were then subsequently annealed to form crystalline iron oxide nanoparticles. The crystalline structure, morphology and magnetic properties of these nanoparticles were investigated using X-ray Diffraction, Scanning Electron Microscopy, Transmission Electron Microscopy, and Physical Properties Measurement System. X-ray Photoelectron and Raman Spectroscopy were also used in order to probe the fine structure and surface characteristics. The biological effect of these nanoparticles was also assessed, using a clonogenic assay for overall cytotoxicity, as well as flow cytometric studies to investigate cellular uptake of the nanoparticles, as well as their effects on the cellular reactive oxygen species (ROS) levels.

While the traditional precipitation method produced roughly spherical hematite particles, the use of spray precipitation resulted in highly porous nanorods. Through XPS and Raman analysis it was found that these nanoparticles feature a small defects of maghemite (γ-Fe2O3) on their surface, in the form of tetrahedrally-coordinated Fe3+. This is due to a a thermodynamically-favoured phase transition on their surface due to being formed from amorphous iron oxide precursors. The concentration of these defects were found to decrease when the precursor was annealed at the higher temperature.

It was found that these defects have an effect on the biological activity, with the spherical nanoparticles showing much lowered toxicity and ROS generation than their rod-shaped counterparts. The particles featuring these maghemite defects, which has been shown to scavenge free radical species, even showed to lower the ROS levels in the cytometric assay relevant to the control. The rod shape of the spray precipitation nanoparticles also had preferential uptake into the MDCK cells compared to the spherical nanoparticles.

This work highlights the need for in-depth understanding of the materials properties of nanomaterials in order to understand the factors governing their toxicity, including effects on their fine structure from synthesis parameters.



Picture2.png

Fate of Various Dendrimers with Different Sizes and Surfaces after Subcutaneous and Intradermal Administration

Thursday, 29th September - 16:00 - Nanomedecine for cancer diagnosis & therapy - Tower 24 - Room 103 - Oral presentation - Abstract ID: 310

Prof. Chie Kojima [1]

1. *Osaka Prefecture University*

Dendrimers are synthetic macromolecules with highly controllable structures, which are potent multifunctional nanoparticles for drug delivery system and imaging. There are many reports on intravenous administration of dendrimer, but few on intradermal and subcutaneous administration. In this study, various dendrimers of different generations (G2, G4, G6, and G8) and different surfaces (amino, carboxyl, acetyl and collagen peptide modification) were prepared and radiolabeled. First, the radiolabeled dendrimers were intradermally administrated to the right footpad of rats. All G2 dendrimers were predominantly accumulated in the kidney. Amino-terminal, acetyl-terminal and carboxy-terminal dendrimers of greater than G4 were mostly located at the injection site, in blood and in the popliteal lymph node, respectively. Interestingly, the carboxy-terminal dendrimers were not largely recognized by macrophages and T cells in the lymph node. Finally, the lymph detection was performed by single photon emission computed tomography (SPECT) imaging using various dendrimers. (Figure 1). Carboxy-terminal dendrimers of greater than G4 and amino-terminal G6 dendrimer (G6-NH2) successfully visualized the popliteal lymph node (indicated by arrows), but acetyl-terminal G6 dendrimer (G6-Ac) and carboxy-terminal G2 dendrimer (G2-COOH) did not. Compared with the COOH-terminal dendrimers of higher than G4, G6-NH2 was largely retained at the injection site (indicated by arrow heads). These results suggest that carboxy-terminal dendrimers of greater than G4 are useful for drug delivery and imaging of sentinel lymph node, which the first lymph node draining tumor cells. Next, the radiolabeled dendrimers were subcutaneously administrated into tumor-bearing mice, and monitored by using SPECT imaging. Acetylated G4 dendrimer (Ac-G4) and collagen peptide-conjugated dendrimer (CP-G4) were largely retained at the injection site for at least 1 day (indicated by arrows). Ac-G4 were partly accumulated in the kidney (indicated by K), but the CP-G4 was not (Figure 2). On the other hand, these dendrimers were accumulated in the liver and the kidney following intravenous administration. These results indicate that the subcutaneously injected dendrimers did not largely gain substantial access to the systemic circulation, which is useful for a depot of drug around the injection site.

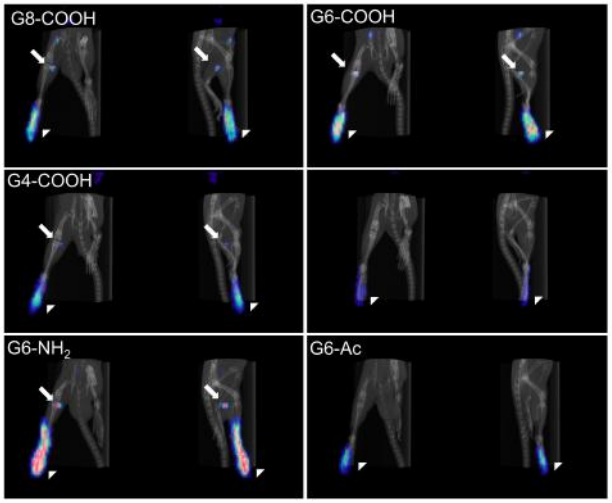


Fig.1.jpg

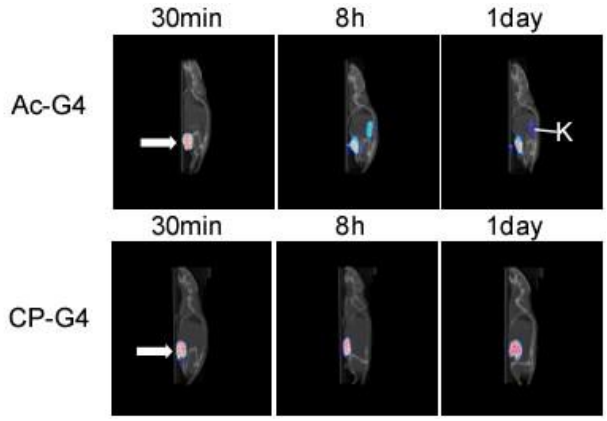


Fig.2.jpg

Specific nanoparticle targeting of the EGF-receptor using single-domain antibodies.

Thursday, 29th September - 16:25 - Nanomedecine for cancer diagnosis & therapy - Tower 24 - Room 103 - Oral presentation - Abstract ID: 46

Dr. Kristof Zarschler [1], ***Dr. Louise Rocks*** [2], ***Dr. Eugene Mahon*** [2], ***Dr. Kanlaya Prapainop*** [2], ***Dr. Holger Stephan*** [1], ***Prof. Kenneth A. Dawson*** [2]

1. *Institute of Radiopharmaceutical Cancer Research, Helmholtz-Zentrum Dresden - Rossendorf,* ***2.*** *Centre For BioNano Interactions (CBNI), School of Chemistry and Chemical Biology, University College Dublin*

Introduction

For effective localization of functionalized nanoparticles at diseased tissues such as solid tumors or metastases through biorecognition, appropriate targeting vectors directed against selected tumor biomarkers are a key prerequisite. The diversity of such oncotropic vector molecules ranges from proteins, including antibodies and fragments thereof, through aptamers and glycans to short peptides and small molecules.

In the presented work we analyze the specific nanoparticle targeting capabilities of a small camelid single-domain antibody (sdAb), representing a potential recognition agent for the epidermal growth factor receptor (EGFR).

Methods

Bioconjugation of EGFR-specific sdAbs to different nanomaterials and characterization of sdAb-conjugates covering in vitro cancer cell imaging, cell proliferation as well as EGFR phosphorylation and signaling are described. The specificity of the sdAb-conjugates is investigated by way of receptor RNA silencing techniques with increasing complexity in vitro by introducing increasing concentrations of human or bovine serum.

Results and Discussion

The results show that sdAb-functionalized nanomaterials can effectively target the EGFR, even in more complex bovine and human serum conditions where targeting specificity is largely conserved for increasing serum concentrations. For highly affine targeting ligands such as sdAbs, targeting a receptor such as EGFR with low serum competitor abundance, receptor recognition function can still be partially realized in complex conditions. Moreover, sdAb-mediated biorecognition of EGFR is not restricted to particular nanomaterials, but was observed to work efficiently in combination with a variety of materials.

Differential Modulation of Biological Properties In Vitro by a Range of cRGDY Peptides on Clinically Translated Dual-Modality Silica Nanoparticles

Thursday, 29th September - 16:50 - Nanomedecine for cancer diagnosis & therapy - Tower 24 - Room 103 - Oral presentation - Abstract ID: 359

Dr. Miriam Benezra [1], ***Dr. Pauliah Mohan*** [1], ***Prof. Ulrich Weisner*** [2], ***Prof. Michelle Bradbury*** [1]

1. *Memorial Sloan-Kettering Cancer Center, New York, NY,* ***2.*** *Cornell University, Ithaca, NY*

Introduction:
Ultra-small Dual Modality nanoparticles, termed Cornell dots (i.e., C' dots) may provide a disease-directed platform for delivery of small cancer molecules for theranostic purposes. For clinical advancement of such platforms is the need to characterize at the cellular and molecular levels, to light biological properties of these probes. We used silica nanoparticle encapsulate Cy 5 dyes (C' dots), surface functionalized with methoxy-terminated polyethylene glycol (PEG), that bind to it different numbers of integrin-targeting cRGDY ligands (N-6, 14, 18) (i.e., cRGDY-PEG-C' dots). Their activities were compared in vitro on several biological systems using cells that over express the αvβ3 receptor, a human melanoma (M21) and human umbilical vein endothelial (HUVEC) cells.
Methods :
Binding and internalization - flow cytometry; Migration performed by time lapse. Adhesion and spreading - SpectraMax5 micro plate reader; Vi-Cell Counter for cell Viability, Effect on signaling - Western blot analysis.
Results:
Dose response and time-dependent binding revealed binding saturation at 50 nM with 14 or 18 cRGDY-PEG-C' dots at two hours as opposed to 100 nM with 6 cRGDY-PEG-C' dots at four hours incubation. Internalization studies revealed a higher uptake (15-20%) of the 14 and 18 particles in 370C compared to 6 cRGDY-PEG-C' dots on M21. Binding specificity was demonstrated using antibodies against the αvβ3 receptor. There was no binding of any of the particle on cells that don't express the αv receptor subunit. Minor binding was observed with two different sizes of scramble peptide-cRADY (6 or 15-20). Additionally, a higher migration rate of HUVEC cells, adhesion with spreading on fibronectin coated plates of M21 cells and proliferation of M21 cells were shown with 14 or 18 cRGDY-PEG-C' dots compared to 6 cRGDY-PEG-C' dots. Activation of the FAK pathway by integrin was also several fold (x2) more with 14 cRGDY as opposed to 6 cRGDY on the particle.
Discussion:
Changes in vitro on the level of binding and key biological processes-cell migration, adhesion/spreading, proliferation process were greater with 14-18 cRGDY-PEG-C' dots. This paves the way in designing a diagnostic platform for primary tumor and cancer metastasis or therapeutic purpose for processes like angiogenesis and wound healing.

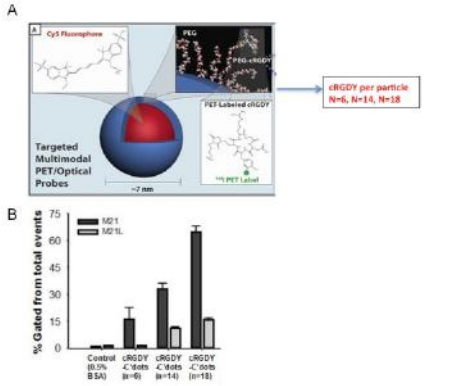


Figure 1: **Multimodal C dot design for $\alpha_v\beta_3$ integrin and characterization** (A) Schematic representation of cRGDY-PEGylated core-shell silica nanoparticle with surface-bearing radiolabels and peptides and core-containing reactive dye molecules (inset). (B) Specific binding of cRGDY-PEG-C dots (N=6,14,18) to M21 cells as against M21-L cells lacking surface integrin expression. Detection of binding was done by flow cytometry. Each bar represents mean±SD of 3 replicates.

Slide3.jpg

Traceable Iron Oxide Based Nanoparticles for Antigen/Adjuvant in vivo Delivery to Lymph Nodes

Thursday, 29th September - 17:15 - Nanomedecine for cancer diagnosis & therapy - Tower 24 - Room 103 - Oral presentation - Abstract ID: 27

Ms. Ane Ruiz de Angulo [1], ***Dr. Aintzane Zabaleta*** [1], ***Ms. Zuriñe Baz*** [1], ***Dr. Jordi Llop*** [1], ***Prof. Juan Carlos Mareque Rivas*** [1]

1. *CIC biomaGUNE*

Most of the currently available vaccines are based on the inactivation of the pathogen, which is not a valid strategy for fighting several chronic infections and, especially, cancer.[1,2] Here we present PEGylated iron oxide based nanoparticles (IONPs) as traceable delivery system for directing tumour antigens and adjuvants to lymph nodes (LNs), leading to improved antitumor immunity and survival outcomes. The nanovehicles allow using lower doses of the immunostimulatory molecules to achieve a given response and thereby enhance their safety profiles. Moreover, other bioactive molecules and drugs can be co-delivered to further improve the efficacy and therapeutic index of the anticancer therapy.

We designed and created PEGylated IONP-filled micelles carrying short oligodeoxynucleotides with CpG motifs as adjuvants and ovalbumin (OVA) as antigen. Doping with rhodamine B-phospholipids[3] was used to monitor in vitro cell uptake, whilst chemistry-free radiolabeling with 67Ga could be applied to study the biodistribution in vivo by SPECT imaging. The immunostimulatory potential of the constructs was tested analyzing cytokine, antibody production and cellular mediated immunity. Immunization against tumour-antigen was performed to demonstrate the anti-tumour immunity.

We obtained water soluble micelles with a diameter below 100nm, which were internalized through the endocytic pathway and co-localized with the toll-like receptor 9 (TLR9). SPECT imaging showed accumulation of these constructs in the LNs and both in vitro and in vivo assays demonstrated enhanced immunostimulatory activity than with the free molecules. The analysis of adaptive immune response demonstrated the production of anti-OVA antibodies and the activation of antigen specific CD8+ T cells, able to significantly slow down the appearance of tumour.

The nanoconstructs were able to deliver the immunostimulatory molecules in the correct intracellular organelle, where the CpGs could reach TLR9 and, therefore, induce cytokine production in vitro. There was no systemic effect in vivo, concluding that the effect was directed to lymphoid organs. The CD8+ T cell subset was successfully activated against the antigen, crucial characteristic for generating anticancer immunity. The anti-tumour studies showed the potential of these systems for developing cancer vaccines.[4]

References

(1)Landrith,T.A. et al. Semin.Immunopathol.2015,37,261–270.

(2)Schreiber,R.D. et al. Science.2011,331,1565–1570.

(3)Cobaleda-Siles,M. et al. Small.2014,10,5054–5067.

(4)Ruiz-de-Angulo,A. et al. ACS Nano.2016,10,1602–1618.

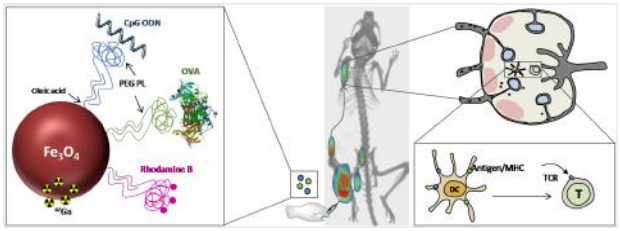

CpG ODN
PEG PL
OVA
Oleic acid
Fe_3O_4
Rhodamine B
^{67}Ga
Antigen/MHC
TCR
DC
T

Figure1.png

Magnetic-nanoparticles as a theranostic tool for liver metastases in a murine model

Thursday, 29th September - 17:40 - Nanomedecine for cancer diagnosis & therapy - Tower 24 - Room 103 - Oral presentation - Abstract ID: 330

Mr. Borja Herrero De La Parte [1], Dr. Eneko Garayo Urabayen [1], Dr. Oihane Kistiñe Arriortua Llarena [2], Dr. Jose Javier Echevarria-uraga [3], Prof. Jose Angel Garcia Martinez [1], Prof. Ignacio García-Alonso Montoya [4], Prof. Fernando Plazaola Muguruza [1], Ms. Irati Rodrigo Arrizabalaga [2]

1. *University of the Basque Country, UPV/EHU,* ***2.*** *BCMaterials,* ***3.*** *Galdakao-Usansolo Hospital,* ***4.*** *University of the*

Introduction: The treatment of liver metastases is still a challenge. Magnetic hyperthermia is being explored as a new therapy. Functionalized magnetic nanoparticles (MNP) can be adhered to tumour tissues and then heated selectively by an alternating magnetic field (AMF). The temperature increment due to the MNP heating of MNP might destroy the tumours spearing healthy liver tissue. The aim of our experiment was to develop a reliable and selective magnetic-hyperthermia therapy for diagnosis and treatment of liver metastases in a rat model. RGD-peptides functionalized MNP, intravascular infusion of MNP and an electromagnetic applicator to achieve selective heating have been designed and an “in vivo” experiment was carried out to check the effectivity of the treatment.

Methods: Colorectal cancer cells were inoculated into the left lateral liver lobe (LLL) of WAG/RijHsd rats. Fe3O4 nanoparticles covered with PMAO and functionalized with RGD peptides were synthetized in order to adhere them to the tumour tissue. Using a microcannula placed into the hepatic artery, MNP-group was infused with MNP and saline-group was infused with the same volume of saline. After 12 hours, MRI studies were practised by 1,5T MRI machine and magnetic hyperthermia was applied by an alternating magnetic field of 14kA/m intensity and 606kHz frequency, during 21 minutes. The temperature was monitored with 3 probes placed in the tumour, LLL and rectum. 12 hours later, both groups were sacrificed and the liver removed for histopathological analysis.

Results: MRI study showed decay of the signal intensity (SI) in the liver and in the periphery of the tumour. Moreover, scattered foci may be observed within the tumours. After thermal exposure, tumour temperature increased 4.66 ± 0.99ºC in saline-group and 7.89 ± 1.2ºC in MNP group ($p<0.001$) (Figure 1). The values of the necrotic tissue, relative to the total tumour tissue, ranged between 1-13% and 13-99% for the saline-group and MNP-group ($p<0.01$), respectively.

Discussion: MNP adhered to liver metastases and could be monitored by MRI. A higher thermal increase in liver and tumour tissue was reached in MNP-infused animals, showing that MNP can be used in magnetic hyperthermia therapy, although further experiments are needed.

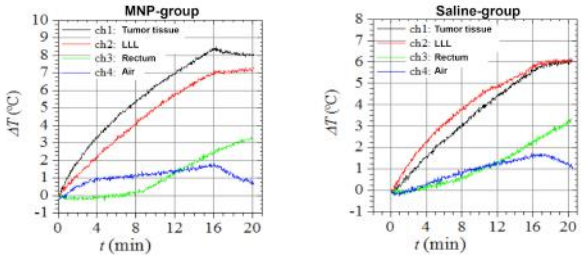

MNP-group
ch1: Tumor tissue
ch2: LLL
ch3: Rectum
ch4: Air
ΔT (℃)
t (min)
Saline-group
ch1: Tumor tissue
ch2: LLL
ch3: Rectum
ch4: Air
ΔT (℃)
t (min)

Figure1.jpg

Hybrids of biomolecules and carbon nanotubes: nanodevices for biosensing

Thursday, 29th September - 16:00 - Biological & medical nanodevices and biosensors - Bionanocatalysis and nanobiosystems - Tower 24 - Room 105 - Oral presentation - Abstract ID: 104

Prof. KAZUO UMEMURA [1], ***Mr. Shusuke Oura*** [1], ***Mr. Yu Ishizaka*** [1]

1. *Tokyo University of Science*

Carbon nanotubes (CNTs) have unique nanostructures and physicochemical properties. For example, when CNTs are irradiated by visible lights, near-infrared (NIR) photoluminescence is observed. On the other hand, when CNTs are irradiated by NIR lights, significant adsorption of NIR is observed. There is another unique phenomenon in fluorescent spectroscopy. When some fluorescent dyes adsorbed on CNT surfaces, fluorescence from the dyes are almost quenched even when the dyes are irradiated by suitable lights for excitation. Using the physicochemical properties, various new methods for nanobiosensing such as detecting single-mismatch of DNA sequences have been reported by various authors.

We have reported nanoscopic characterization of hybrids of DNA and CNTs (DNA-CNT hybrids) in order to obtain fundamental information of interaction between DNA and CNTs. [1-6] Furthermore, we have investigated reactions of DNA binding proteins with DNA-CNT hybrids to establish biochemical reactions on CNT surfaces.

In this paper, we systematically studied optical responses of DNA-CNT hybrids under various conditions. UV-vis, NIR, photoluminescence, and fluorescent spectroscopies were applied to the same samples for comparison. As a result, we found that DNA-CNT hybrids can detect changes of the spectra caused by minute perturbation such as injection of chemicals. Our results will be a basis of application of CNTs to nanobiosensing. In addition, causative proper use of the several optical measurements dependent on research purposes will be discussed.

[1] T Hayashida and K Umemura, Colloids Surfaces B, 143, 526-531(2016).
[2] K Umemura. Nanomaterials, 2015, 5(1), 321-350(2015).
[3] S Oura, M Ito, D Nii, Y Homma, K Umemura. Colloids Surfaces B, 126, 496–501(2015).
[4] D Nii, T Hayashida, Y Yamaguchi, S Ikawa, T Shibata, K Umemura, Colloids Surfaces B, 121, 325-330(2014).
[5] D Nii, T Hayashida, K Umemura. Colloids Surfaces B, 106, 234–239(2013).
[6] T Hayashida, K Umemura. Colloids Surfaces B, 101, 49-54(2013).

Biomimetic compartmentalization approach in designing nanoreactor with organelle-like function

Thursday, 29th September - 16:25 - Biological & medical nanodevices and biosensors - Bionanocatalysis and nanobiosystems - Tower 24 - Room 105 - Oral presentation - Abstract ID: 274

Dr. Vimalkumar Balasubramanian [1], ***Ms. Alexandra Correia*** [1], ***Dr. Hongbo Zhang*** [1], ***Ms. Flavia Fontana*** [1], ***Mr. Ermei Mäkilä*** [2], ***Prof. Jarno Salonen*** [2], ***Prof. Jouni Hirvonen*** [1], ***Dr. Helder A. Santos*** [1]

1. *University of Helsinki,* ***2.*** *University of Turku*

Introduction

In nature, biological cell is regarded as an intricated microenvironment, in which myriad of enzymes work together to catalyze concurrently multiple chemical reactions concurrently that are crucial for metabolism and cell functions. Inspired by nature, scientists manifest an increasing interest to mimic these compartmentalized enzyme regulatory mechanisms, not only to understand the biological process of cellular metabolism and functions, but most importantly to design customized biomimetic nanomachineries with a tremendous potential in a range of applications from biochemicals, pharmaceutics, diagnostics to, biomedicine, smart materials, and synthetic organelles. Therefore, mimicking the chemical transformations exhibited by enzymes in cellular compartments has triggered intensive research interest for creating new dynamic materials with tunable enzyme reactivity. In this work, we aim to develop a compartmentalized cellular nanoreactor consisting of porous silicon nanoparticles (PSi NPs) entrapped with an enzyme, horseradish peroxidase (HRP), and surface coated with a cancer cell membranes to demonstrate the design of biomimetic cellular nanoreactors and their impact on improving cellular functions for biomedical applications.

Methods

PSi NPs have been used as host material to the entrapment of HRP enzymes within the pores. To construct a compartmentalized cellular nanoreactor, purified cancer cell membranes coated the surface of PSi NPs by an extrusion. The enzyme activity and kinetics of the compartmentalized cellular nanoreactors were evaluated using an enzymatic assay. To demonstrate the function of biomimetic cellular nanoreactors in improving cellular functions, different in-vitro cell experiments were performed.

Results and Discussion

The results from the physicochemical characterization of nanoreactors indicate the successful loading of HRP in PSi NPs and the cancer cell membrane surface coating. The enzyme activity and kinetics analyses showed an enhanced substrate affinity and reaction rate compared to free HRP (Figure 1B), suggesting a high catalytic activity of nanoreactors. The in-vitro cell experiments demonstrated that nanoreactors can readily integrate with cells and can supplement the cellular functions under oxidative stress conditions. This cellular nanoreactor featuring a biocompartment enclosed by cellular membrane closely resembling nature's biocompartmentalization is an innovative biomimetic approach, which further helps the customized designing of biomimetic nanomachineries that can impact the repairing of cellular functions in pathological conditions.

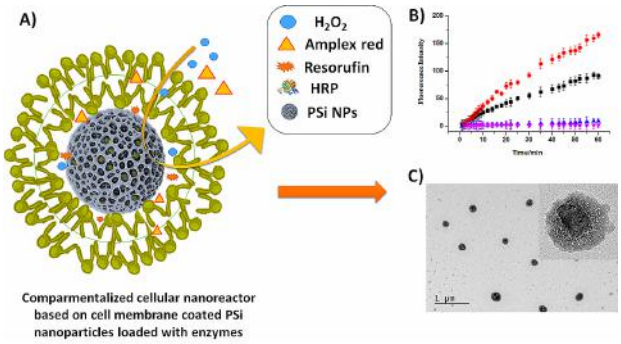


Figure 1. *A) Schematic representation of the biomimetic compartmentalized nanoreactor based on the cancer cell membrane coated PSi NPs loaded with HRP enzymes. B) Enzyme activity of nanoreactors analyzed using amplex-red fluorescence assay, compared with different controls. C) Transmission electron micrograph of the compartmentalized cellular nanoreactors.*

Figure 1.png

NBelyax® Nanoparticles for disinfection and sterilization of living and nonliving surfaces tested in vitro and in situ

Thursday, 29th September - 16:50 - Biological & medical nanodevices and biosensors - Bionanocatalysis and nanobiosystems - Tower 24 - Room 105 - Oral presentation - Abstract ID: 426

Dr. Leon Albarran [1], ***Mrs. Gabriela León*** [1], ***Mr. Sergio León*** [1], ***Dr. Paola Arteaga*** [1]

1. *Gresmex S.A. de C.V.*

Gresmex is a Mexican company with 17 years of experience in manufacturing cleaning products and personal care.

In this project the active nanoparticle NBelyax® was developed. The invention of this material began in 2008 as part of the research enterprise environment to a material or substance capable of removing viruses.

In the first stage of innovation we worked on the elimination of rotavirus considering it as the most resistant virus to external factors, and that it could serve as a reference for the removal and inactivation of any other type of virus. The second stage was focused on the evaluation of effectiveness against a wider spectrum of microbiological organisms, including viruses, bacteria, fungi, spores, mycobacteria and trypanosomes. The mechanism of action is theoretically based on the nanoparticle attacking the genetic material of pathogenic microorganisms.

The Bioselectivity of the active ingredient NBelyax® gives us the advantage over other developments or products to eliminate all pathogens without causing any damage to the cells or the body of the user. Moreover, protect against infections supports better healing wounds existing prior to use.

The third stage was the inclusion in the portfolio of finished antiseptics, disinfectants and sterilants, in order to give a high disinfecting power and therefore a high added value to our products. That was the birth of the Eviter® products which include sanitizers, soaps, creams and cold sterilants designed for use in medical, clinical and surgical areas

NBelyax®, being broad spectrum disinfectant and high level is an important tool for prevention of diseases caused by active infections.

It is noteworthy that Eviter® can be used in any environment to create biosecure environments such as schools, museums, offices, transportation and avoid contagion large vectors that trigger pandemics.

Cloning, expression and purification of Pseudomonas aeruginosa azurin, a small redox protein and its application in molecular electronics

Thursday, 29th September - 17:15 - Biological & medical nanodevices and biosensors - Bionanocatalysis and nanobiosystems - Tower 24 - Room 105 - Oral presentation - Abstract ID: 22

Ms. Neeti Kalyani [1], ***Prof. Prashant Mishra*** [1]

1. *IIT Delhi*

INTRODUCTION

Redox metalloproteins are getting considerable interest nowadays due to their applications in creating hybrid sub micrometre-sized electronic components for biosensors, transistors and memory devices. These bionanodevices overcome the limitations of silicon-based devices like tunnelling effects, efficiency, and heat. These are operationally simple, cost efficient and suitable for real time detection. Azurin from Pseudomonas aeruginosa is an oxidoreductase, which possess an inherent, efficient electron transfer capability, occurring at single electron level in a very fast and directional way. Also, this has been proved to stabilize p53 tumor suppressor protein and hence, useful in cancer therapy. Azurin has a great potential to be used in the molecular electronics. But the purification of this protein is difficult and require multiple steps which result in low yield. We have cloned the protein and purify it in single step and got better yield of the protein. We have confirmed its electron transfer properties with cyclic voltammetry and conductive AFM.

METHOD

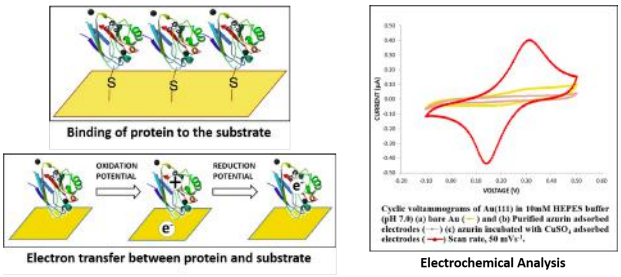
Azurin gene was cloned and expressed in Escherichia coli and purified by Ni-NTA (Nickel-Nitrilo acetic acid) chromatography. Recombinant azurin was characterized using UV-Visible, FTIR, Circular Dichroism and Raman Spectroscopy. Azurin was immobilized directly on gold via thiol linkage and on silicon and mica using 3-mercaptopropyl trimethoxysilane as linker. Conductive atomic force microscopy was also done to study the morphology and conductivity of azurin layer on different substrates.

RESULTS AND DISCUSSION

Azurin gene was successfully cloned and expressed in good amount. Pure band of azurin was obtained after single step of Ni-NTA purification. The excellent current density of azurin layer on gold was observed in cyclic voltammetry experiments. The sharp oxidation and reduction potentials peaks were as reported for the wild type azurin. In conductive atomic force microscopy the current observed was higher in the azurin immobilized sample than the blank one.

CONCLUSION

We were successful in getting the pure active protein in very good quantity which can be used directly for electronic studies. Same protein can be used as the acceptor and donor of electrons. Its small size and efficient electron transfer capability make it a desirable molecule for the development of biosensors.



Azurin protein for biosensor.png

A modified Hodgkin–Huxley model for nanoelectronics

Thursday, 29th September - 17:40 - Biological & medical nanodevices and biosensors - Bionanocatalysis and nanobiosystems - Tower 24 - Room 105 - Oral presentation - Abstract ID: 392

Prof. Peter Burke [1]

1. *University of California, Irvine*

A modified Hodgkin–Huxley model for nanoelectronics

In the Hodgkin–Huxley model[1], bioligcal ion pumps push current through a membrane until is the electrical capacitance of the membrane is charged up, at which point it generates a voltage which turns off the ion channel pumps. When a lipid bilayer is brought into close proximity to a nano-electronic device (such as a 1d carbon nanotube[2], quasi-1d silicon nanowire, 2d graphene sheet[3], or any other of the many classes of nanoelectronics devices), a similar phenomenon occurs: The current through the ion channel charges up the electrical capacitance of the lipid bilayer. However, because the nano-electronic devices have capacitance of their own (including both classical electrical capacitance as well as quantum capacitance[4] due to the Pauli exclusion principle), these also get charged by the ion channels, and ultimately turn off the ion channels, even for simple ion channels that are not voltage dependent. This modified Hodgkin–Huxley model can account for the electrical behavior of nanoelectronic interfaces to a electrically active ion channels, a growing interest with applications from neurons and cadiomyoctyes to even smaller, electrophysiologically active organelles such as chlorplasts and even mitochondria[5].

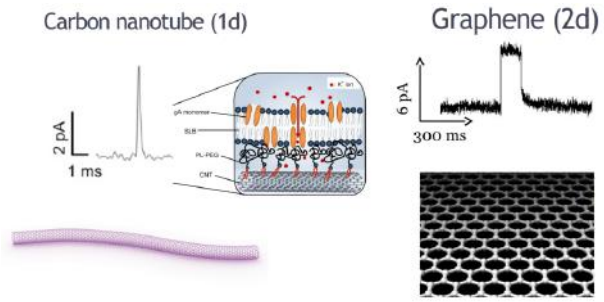
[1]A. L. Hodgkin and A. F. Huxley, “A quantitative description of membrane current and its application to conduction and excitation in nerve,” J. Physiol., vol. 117, no. 4, pp. 500–544, Aug. 1952.

[2]W. Zhou, Y. Y. Wang, T.-S. Lim, T. Pham, D. Jain, and P. J. Burke, “Detection of single ion channel activity with carbon nanotubes,” Sci. Rep., vol. 5, p. 9208, 2015.

[3]Y. Y. Wang, T. D. Pham, K. Zand, J. Li, and P. J. Burke, “Charging the quantum capacitance of graphene with a single biological ion channel.,” ACS Nano, vol. 8, no. 5, pp. 4228–4238, Apr. 2014.

[4]C. Rutherglen and P. Burke, “Nanoelectromagnetics: Circuit and Electromagnetic Properties of Carbon Nanotubes,” Small, vol. 5, no. 8, pp. 884–906, 2009.

[5]K. Zand, T. Pham, A. Davila Jr., D. C. Wallace, and P. J. Burke, “Nanofluidic platform for single mitochondria analysis using fluorescence microscopy,” Anal Chem, vol. 85, no. 12, pp. 6018–6025, 2013.



Screenshot from 2016-07-15 13 28 01.png

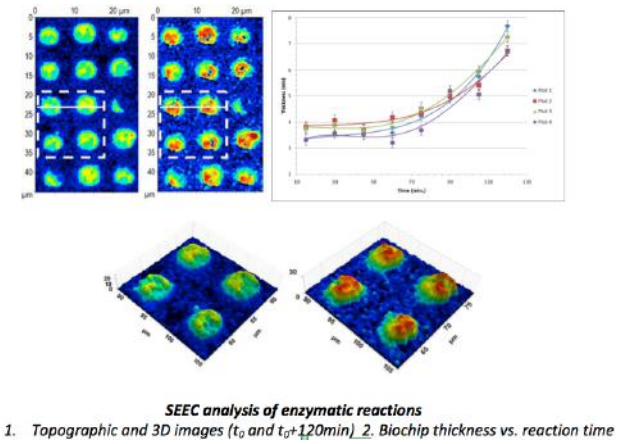
WORKSHOP: SEEC Microscopy, a live and label-free analysis technique in the fields of Materials and Life Sciences

Thursday, 29th September - 16:00 - Nano-Imaging for diagnosis, therapy and delivery - Tower 24 - Room 107 - Workshop presentation - Abstract ID: 574

Mr. Nicolas Medard [1], *Mr. Imed Ayadi* [1]

1. *Nanolane*

SEEC Microscopy is a label-free analysis technique offering new characterization capabilities such as the live nanoscale imaging, the multiplex molecular interaction analysis or the real-time quantitative study. The technique implements unique optical sensitive sensors (SEEC sensors) with specific contrast-enhanced properties enabling the live visualization of samples down to nanoscale (0.1nm). In addition, a proprietary algorithm (Q-SEEC) enables quantitative analyses (surface interaction and topography analyses) with an accuracy of 0.3nm.
SEEC Microscopy is dedicated to the study of samples in the fields of Materials and Life Sciences. Thanks to its high lateral sensitivity (down to 1 nm), the technique can be applied for the analysis of nanofilms, nanopatterns, nanotubes, nanoparticles or DNA molecules… Successful analyses were recently performed on polyelectrolytes, lipid layers, biofilms, biochips samples.
The N-Lab Station, brand-new SEEC equipment, is commercializedin two versions: the 'MAT' version (for analyses in dry) with a precise thermal control stage and the 'BIO' version (for analyses in dry and liquid) including complete thermal and fluidic systems (injection pump, fluidic switch, fluidic chamber, flowmeter…).
Bothversions are providedwith the 'LabSoft' software for sequence planning, live data display and treatment.
Moreinformation: `http://nlab.nano-lane.com/`
SEND AN EMAIL TO nicolas.medard@eolane.com TO SECURE YOUR SEAT FOR THIS WORKSHOP.



Seec analysis of enzymatic reactions.png

N-lab station.png

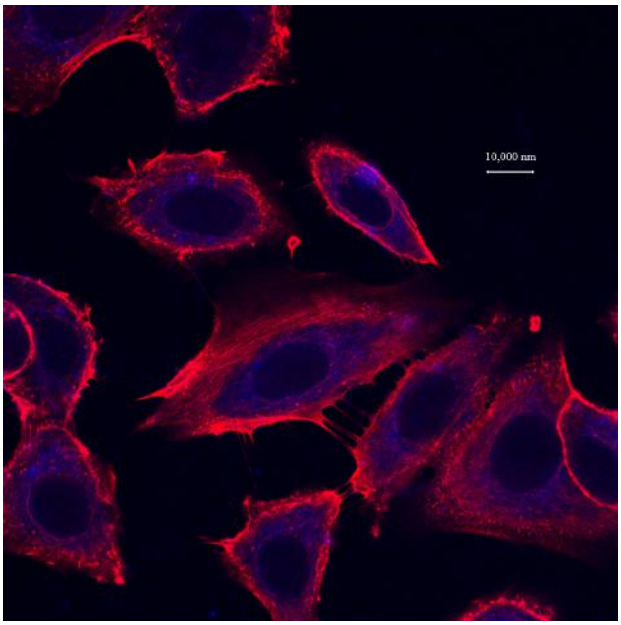
Functionalization of Emissive Conjugated Polymer Nanoparticles by Coprecipitation: Consequences for Particle Photophysics and Colloidal Properties

Thursday, 29th September - 16:45 - Nano-Imaging for diagnosis, therapy and delivery - Tower 24 - Room 107 - Oral presentation - Abstract ID: 523

Prof. Gareth Redmond [1]

1. *University College Dublin*

Recent developments in materials synthesis, bio-conjugation methods and luminescence techniques have led to a rapid proliferation of novel fluorescence-based approaches to imaging in the life sciences. However, for high-resolution or long-time duration imaging applications, molecular dyes suffer from limitations of low brightness, poor photo-stability and fluorescence intermittency (blinking). Consequently, we are exploring a potentially promising alternative based on highly fluorescent conjugated polymer materials. Here, the functionalization of polyfluorene (PFO) nanoparticles by coprecipitation of the conjugated polymer with an amphiphilic comb polymer, consisting of a hydrophobic polystyrene backbone with hydrophilic, carboxylic acid-terminated polyethylene oxide side-chains (PS-PEG-COOH), is investigated. The comb polymer affects the properties of the formed hybrid nanoparticles. Non-functionalized particles are typically larger (28 nm) than functionalized ones (20 nm); this size difference impacts peak molar extinction coefficients accordingly. Zeta potentials are negative, consistent with negative surface charge on PFO particles due to chemical defect formation, with additional charge on functionalized particles due to the pendant carboxylic acid groups. Emission quantum yields of functionalized particles are typically larger, consistent with lower efficiency of energy transfer to quenchers in smaller particles and weaker PFO interchain interactions due to chain dilution. The trend in per-particle fluorescence brightness values, as confirmed by single particle fluorescence imaging, reflects the size-dependent nanoparticle absorption cross sections. Photostability studies on aqueous dispersions of hybrid particles indicate mild photobrightening under continuous illumination while PFO particles exhibit slow exponential emission decay. Functionalized particles also tend to be more resistant to aggregation during exposure to adenocarcinoma cells; see Figure (polyfluorene nanoparticles (blue) in MCF-7 cells (red / phalloidin-TRITC)). Generally, the hybrid particles exhibit more favourable time-, pH- and medium-dependent stabilities, likely due to steric and electrostatic stabilization by PEG- carboxylic acid functionalities. Overall, the functionalized particles exhibit attractive properties: Reasonably small size, tight size distribution, high absorption cross section, radiative rate and emission quantum yield, excellent brightness and photostability, and good colloidal stability.



Mcf-7sample7image3series z07c1 2.jpg

Innovative SPIONs for multimodal imaging: MRI/PET and MRI/optical imaging

Thursday, 29th September - 17:10 - Nano-Imaging for diagnosis, therapy and delivery - Tower 24 - Room 107 - Oral presentation - Abstract ID: 13

Dr. Julien Boudon [1], Dr. Guillaume Thomas [1], Dr. Lionel Maurizi [1], Prof. Nadine Millot [1]

1. *Université Bourgogne Franche-Comté*

Amongst non-invasive methods, single photon emission computed tomography (SPECT), positron emission tomography (PET) and magnetic resonance imaging (MRI) are routinely used to detect pathological tissues. Each of these imaging modalities has unique advantages along with intrinsic limitations (for example MRI has an excellent spatial resolution but suffers from a lack of sensitivity). To circumvent these hurdles, ongoing efforts are made to develop multimodal methods. Similarly, bimodal medical imaging relying on MRI coupled to Optical Imaging (OI) may improve the accuracy of a diagnosis by combining the sensitivity of OI and the resolution of MRI.

In this field, nanoparticle (NP) technologies represent the most promising strategy to elaborate multimodal contrast agents. Superparamagnetic iron oxide nanoparticles (SPIONs) are currently being developed for several biological applications such as hyperthermia, drug delivery, or MRI [1-2]. For the latter application, SPIONs are usually used as water proton transverse relaxation time (T2) contrast agents.

The SPIONs presented in this talk for multimodal imaging (MRI/PET and MRI/optical imaging) are developed by taking advantage of (i) the originality of their simultaneous synthesis and colloidal stabilization in one step through a continuous hydrothermal synthesis device (synthesis time <10 s, conditions T, P up to the supercritical water field) [3-4] and (ii) the association of these nanoparticles with innovative chelating agents (NODAGA, MANOTA etc.) [5] or innovative fluorophores like phthalocyanine derivatives for near-IR detection and photodynamic therapy [6].

For all the nanohybrids developed, the grafting of the different ligands is confirmed by exhaustive characterizations. The imaging agents are stable under physiological conditions and show no cytotoxicity. Yields of radiolabeling indicate an efficient chelation and subsequent spectroscopic studies address the imaging capabilities of each probe in the well characterized SPIONs. In vitro, in vivo and biodistribution results of the final NPs will be presented.

[1] L. Maurizi et al., Langmuir, 2009, 8857

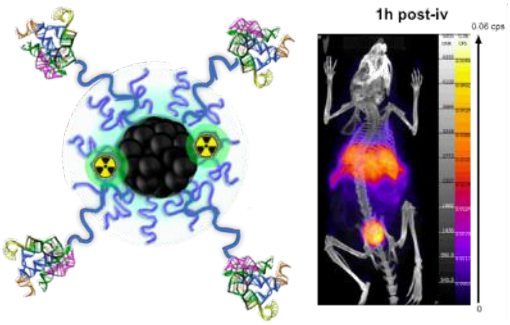
[2] L. Maurizi et al., J. Biomed. Nanotechnol. 2015, 126

[3] L. Maurizi et al., Chem. Commun., 2011, 11706

[4] G. Thomas et al., RSC Adv., 2015, 78614

[5] G. Thomas et al., Nanotechnology, 2016, 135604

[6] J. Boudon et al., Chem. Commun., 2013, 7394



Iconan2016 spions image.png

Elaboration of a new in vivo imaging system based on multimodal upconversion nanoparticles

Thursday, 29th September - 17:35 - Nano-Imaging for diagnosis, therapy and delivery - Tower 24 - Room 107 - Oral presentation - Abstract ID: 410

Mr. Julien Santelli [1]***, Dr. Lechevallier Séverine*** [2]***, Dr. Robert Mauricot*** [3]***, Prof. Daniel Cussac*** [4]***, Prof. Marc Verelst*** [5]

1. *CEMES-CNRS, Université de Toulouse, CNRS, France | I2MC, Université de Toulouse, INSERM, Hôpitaux de Toulouse, France,* ***2.*** *CHROMALYS SAS, 29 rue Jeanne Marvig BP 94347 31055 TOULOUSE cedex 4, France,* ***3.*** *CEMES-CNRS, Université de Toulouse, CNRS, France,* ***4.*** *I2MC, Université de Toulouse, INSERM, Hôpitaux de Toulouse, France,* ***5.*** *CEMES-CNRS, Université de Toulouse, CNRS, France | CHROMALYS SAS, 29 rue Jeanne Marvig BP 94347 31055 TOULOUSE cedex 4, France*

Background

In vivo imaging can be achieved with various techniques (MRI, CT-scan, PET-scan, scintigraphy, fluorescence…) depending on the nature of the target tissue, cell or tracer. Nevertheless, there is actually no reference method that allows for optimal results: each technique has advantages and limitations in depth, resolution, sensitivity, tissue damage or cost.

Methods

We have developed multimodal nanoparticles for biological imaging: a Gadolinium oxysulfide core allows MRI and CT-scan imaging whereas the addition of other lanthanides as dopants enables the acquisition of fluorescence properties. The combination of the different techniques gives maximal information with only one tracer. This brings much more flexibility and robustness in the results comparatively to other tracers currently available on the market. We have found that Ytterbium/Thulium codoped nanophosphors are particularly suited for in vivo imaging as they emit strong luminescence after upconversion of near infrared photons. Briefly, this enables the imaging of deeper targets whilst minimizing tissue lesions compared to classical fluorescence obtained after visible laser excitation. Indeed, it benefits from tissue transparency and lower excitation energy.

Findings

Here we report the development of a complete in vivo fluorescence tracking system: Ytterbium/Thulium codoped Gadolinium oxysulfide as tracer and dedicated macroscopic imaging device. Our latest results are very promising: we are able to follow the kidney-injected tracer on the surface of the skin after two months.

This let us foresee real applications to achieve our final goal: over-time detection of nanoparticle-labeled mesenchymal/stromal cells after grafting in a solid organ. In the end, it will signify the availability of an innovative and robust tool for cell therapy proof of concept.

The Curious Case of 1D and 2D Carbon Nanostructure Pharmacology & Toxicology

Friday, 30th September - 09:00 - Plenary Speeches - Amphitheatre 25 - Oral presentation - Abstract ID: 573

Prof. Kostas Kostarelos [1]

1. *University of Manchester*

Carbon nanomaterials may have captured the excitement and interest of myriads of scientists and two Nobel prizes, but have only just entered a phase of maturity in their development and utilization for medical purposes. Graphene-based materials that constitute some of the newest nanocarbon types today, have mainly been explored as components of biosensors and for construction of matrices in tissue engineering. The capacity of graphene to act as a platform for various therapeutic and diagnostic agents has also been reported, however not as coherently. Among the recent advances made with graphene-related materials, their use as components of innovative delivery systems is full of promise, however there are challenges facing these exciting new tools both in terms of biological activity and toxicological profiling. This presentation will attempt to offer a comparative perspective on the pharmacological and toxicological profiles revealed for 1D (carbon nanotubes, CNT) and 2D (graphene oxide sheets, GO) carbon nanostructures. This aims to be an illustration of the importance rational and systematic research work plays to unravel the unparalleled properties of carbon nanomaterials and allow their utilization as clinical tools.

Nucleic acid chemistry for nanomedicine

Friday, 30th September - 09:40 - Plenary Speeches - Amphitheatre 25 - Oral presentation - Abstract ID: 526

Prof. philippe Barthelemy [1]

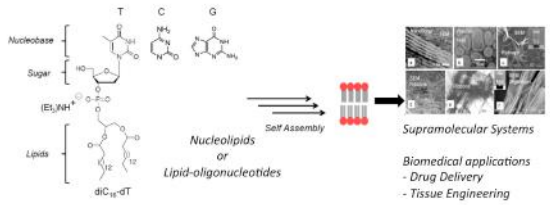
1. *INSERM U1212*

The combination of nucleic acids chemistry (e.g., nucleoside, nucleotides, oligonucleotides) with supramolecular principles provides an efficient and powerful approach to prepare well-defined systems with tunable physico-chemical properties and functions. We develop new nano-systems based on nucleic acids chemistry for i) drug delivery applications (therapeutic, theranostic), and ii) tissue engineering. This communication will present novel “smart” nucleic acid derivatives (nucleolipids, lipid-oligonucleotide conjugates) developed in our lab [1].

References

[1]

- Latxague, L., Ramin, M.A., Appavoo, A., Berto, P., Maisani, M., Ehret, C., Chassande, O., and Barthélémy, P. (2015) Angewandte Chemie, DOI: 10.1002/ange.201409134.
- Oumzil, K. Benizri, S. Tonelli, G. Staedel, C. Appavoo, A. Chaffanet, M. Navailles L. and Barthélémy P. (2015) ChemMedChem DOI: 10.1002/cmdc.201500381
- Luvino, D. ; Khiati, S. ; Oumzil, K. ; Rocchi, P. ; Camplo, M. ; and Barthélémy P. (2013) J. Control. Release, 172, 954–961
- Khiati, S., Luvino, D., Oumzil, K., Chauffert, B., Camplo, M. and Barthélémy P. (2011) ACS Nano, 5 (11), pp 8649–8655.
- Patwa, A., Gissot, A., Bestel, I. and Barthélémy, P. (2011) Chem. Soc. Reviews, 40, 5844–5854.
- Godeau, G., Bernard, J., Staedel, C., Barthélémy, P. (2009) Chem. Commun., 5127-5129.
- Moreau, L., Camplo, M., Wathier, M., Taib, N., Laguerre, M., Bestel, I., Grinstaff, M.W., Barthélémy, P. (2008) J. Am. Chem. Soc. 130, 14454-14455.



Diapositive1.jpg

Nanotechnologies for targeted delivery of nucleic acid

Friday, 30th September - 10:45 - Plenary Speeches - Amphitheatre 25 - Oral presentation - Abstract ID: 517

Prof. Elias Fattal [1]

1. *Université Paris-Sud*

Hyaluronic acid (HA) is a glycosaminoglycan, the main constituent of the extracellular matrix and the natural ligand of CD44 receptor. The association of HA with nanotechnology allow to target the cancer stem cells through the CD44 receptor overexpressed on the surface of these cells. Lipoplexes containing a HA-dioleoyl phosphatidylethanolamine conjugate (HA-DOPE) were designed for this purpose. They were prepared from cationic liposomes and used to complex small interfering RNA (siRNAi). Targeting the CD44 receptor on lung cancer cells was shown to improve the inhibition effect of siRNA using the luciferase gene as a target. The internalization mechanism of the lipoplexes was shown to be mediated by both the CD44 receptor and caveolae. This approach has been applied successfully to deliver the same siRNA anti-luciferase in a mouse model of lung metastases demonstrating a higher inhibition than the non-targeted formulation. We have also associated an aptamer antiCD44 to the surface of liposomes. These vectors have also shown great potential for siRNA delivery in vitro and in vivo to cells overexpressing CD44 avoiding the toxicity problems related to lipoplexes. These data validates the relevance of CD44 as a target for the delivery of macromolecular drugs by nanotechnology. Finally, we have designed inhaled formulation for siRNA delivery in the treatment of lung injury disease, to reduce inflammation. Dendriplexes delivery of an antiTNF siRNA have shown, after intratracheal administration, a potent inhibition of this cytokine demonstrating the potential of nanotechnologies to also deliver locally the siRNA.

Nanostructured Biomaterials for Medical and Biological Applications

Friday, 30th September - 11:25 - Plenary Speeches - Amphitheatre 25 - Oral presentation - Abstract ID: 558

. Jackie Ying [1]

1. *Institute of Bioengineering and Nanotechnology*

Nanostructured materials have been developed for various medical and biological applications. They have been designed as stimuli-responsive drug delivery systems and sustained protein delivery systems. Nanocomposite systems have also been derived to provide simultaneous drug delivery and bioimaging functions as theranostic systems. Micellar nanocomplexes have been synthesized with green tea-based ingredients as unique carrier materials that offer synergistic therapeutic effects with the drugs to be delivered.
In addition, nanostructure processing has been employed in creating synthetic cell culture substrates for the expansion and controlled differentiation of stem cells. Nanostructured scaffolds have also been obtained for cell and tissue engineering.

Investigating the Eremostachys laciniata (EL) and Curcumin Longa (CL) encapsulated by Solid Lipid Nanoparticles (SLN) in order to treat the inflammatory diseases

Friday, 30th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 76

Mr. akbar vaseghi [1], Ms. Haleh Seyedabasi [2], Ms. Mina Ghanbari [3], Dr. Alireza Panahi [4], Ms. Elaheh Alizadeh [5], Mr. Bager Karimi [6], Mr. Reza Ashrafi Parchin [7]

1. *Department of Genetics, Faculty of Biological Sciences, Tarbiat Modares University, Tehran, Iran,* ***2.*** *Department of Agriculture, Khoy Payamnor University, Western Azerbaijan, Iran,* ***3.*** *Department of Applied Chemistry, Faculty of Science, University of Mohaghegh Ardabili, Ardabil,* ***4.*** *Department of Biology, Faculty of Basic Sciences, University of Mohaghegh Ardabili , Ardabil, Iran,* ***5.*** *Department of Biotechnology, Payame Noor University , Tehran,* ***6.*** *Department of Biotechnology, Faculty of Agriculture, Tarbiat Modares University, Tehran, Iran,* ***7.*** *Department of Biotechnology and Plants Breeding, Gorgan University of Agricultural Sciences and Natural Resources, Iran*

Solid Lipid Nanoparticles (SLN) have emerged as a next-generation drug delivery system with potential applications in pharmaceutical field, cosmetics, research, clinical medicine and other allied sciences. Since a decade, trials are being made to utilize SLN as alternative drug delivery system to colloidal drug delivery systems such as lipid emulsions, liposomes and polymeric nanoparticles. Eremostachys laciniata(EL), an Iranian traditional medicinal herb, and Curcumin Longa (CL) are a thick root which are used in local traditional medicine in order to treat the inflammatory diseases. In this project, we are studying nano-encapsulation by using high-pressure homogenizer method with EL and CL effective material mixtures, isolated from rhizomes, and SLN (we call NANO-ChiCo). We compared the NANO-ChiCo with piroxicam ointment in order to treat the inflammatory diseases, e.g., osteoarthritis, rheumatoid arthritis, and reiter's syndrome. After one month of treatment with the SLN-ChiCo and piroxicam ointment, our recent results for all groups showed significant improvement compared to the control groups.

Effects of paclitaxel delivery by carbon nanotubes on prostate cancer cells and monocytes

Friday, 30th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 85

Mr. Edson Comparetti [1]***, Dr. Valber Pedrosa*** [2]***, Dr. Ramon Kaneno*** [1]

1. *Institute of Biosciences of Botucatu, UNESP - Univ Estadual Paulista, Department of Microbiology and Immunology,* ***2.*** *Institute of Biosciences of Botucatu, UNESP - Univ Estadual Paulista, Department of Chemistry and Biochemistry*

Carbon nanotubes (NT) have properties that make them useful in the biomedical field, for instance for controlled drug delivery. Indeed, NT can be loaded with antitumor drugs, such as paclitaxel (PTX), in order to guide drug release within the tumor cells. Considering the high-energy metabolism of tumor cells, we revested PTX-loaded NT with glucose molecules (GLY) in order to test their toxicity on both human prostate cancer cells and monocytes of health donors. First, we increased the polydispersity of NT in aqueous solution synthesizing them with polyethyleneimine (PEI) and polyethyleneimine combined with glucose (PEI-GLY). Analysis by Fourier transform infrared spectroscopy (FTIR – Nicoletspectrometer Nexus 670) confirmed the incorporation of polymer on NT walls. Transmission electron microscopy was used to study NT structure and morphological changes induced by incorporated material. The amount of PTX incorporated by NT was estimated by UV/visible spectroscopy. Cytotoxic activity on prostate cancer cells (LNCaP) was confirmed by flow cytometry after 48h incubation with nanoparticles. Analysis of cell viability was based on Annexin V- (apoptosis) and 7AAD- (cell death) labeling and analysis by flow cytometry in five experimental conditions: a) NT; b) NT-PTX; c) NT-GLY; d) NT-GLY-PTX and e) equivalent concentration of pure PTX. NT-PTX showed as high cytotoxic activity as pure PTX, that was significantly higher than NT alone or NT-GLY. The most interesting finding was that cytotoxic effects of NT-GLY-PTX had slightly higher than pure PTX and NT-PTX, indicating that glucose favors the interaction of particles with tumor cells (Fig.1). We also analyzed the effect of NT on the viability and phenotype of monocytes of health donors exposed to NT, NT-PTX, NT-GLY and NT-GLY-PX. Our results show that expression of CD80, CD86, HLA-DR and CD83 was not hindered by exposition to nanotubes (any of preparations), while all formulations increased the expression of PD-L1 molecules on monocytes. These results indicate that glucose enhance the drug-carrier property of NT, and has low toxic effect on normal monocytes.

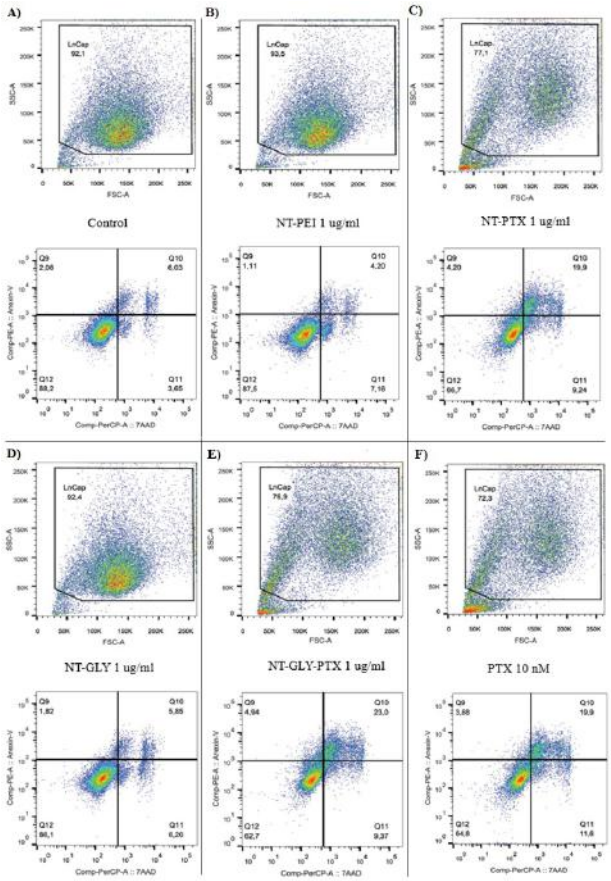


Fig. 1 - Effects of modified carbon nanotubes on morphology and viability of tumor cells. Dot plot depicts the effect of NT-PTX, NT-GLY-PTX and pure PTX (C, E and F) on the size and granulocyte of prostate cancer cell line LNCaP. Pseudocolor graphs represent early apoptosis (Annexin V+, Q9), late apoptosis (Annexin V+/7AAD+ cells, Q10) and dead cells (7AAD+ cells, Q11).

Cell death.jpg

Nanoscale Engineering of Hybrid Magnetite-Carbon Nanofibre Materials for MRI Contrast Agents

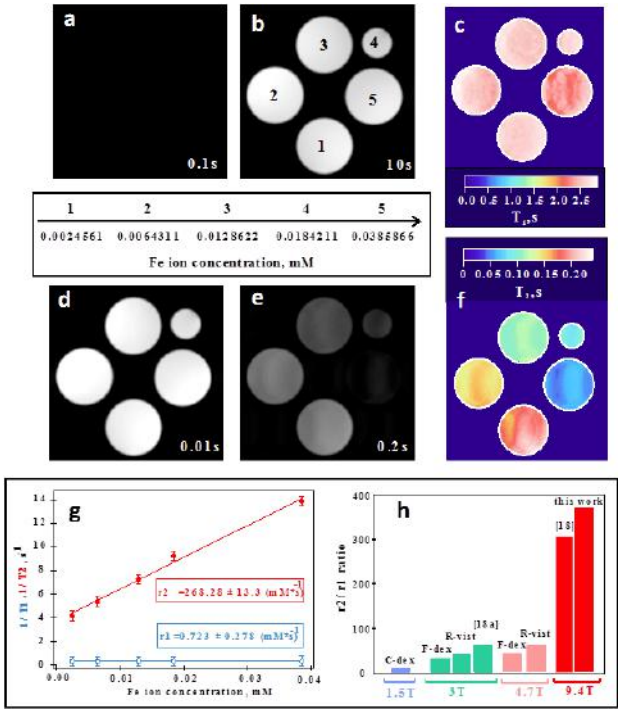
Friday, 30th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 111

Ms. Olga Metelkina [1], ***Dr. Graham Rance*** [2], ***Dr. Galina Pavlovskaya*** [2], ***Prof. Alexander Savchenko*** [1], ***Prof. Andrei Khlobystov*** [2], ***Prof. Alexander Majouga*** [1], ***Dr. Anastasia Garanina*** [1]

1. *National University of Science and Technology, MISiS,* ***2.*** *The University of Nottingham*

Magnetic nanomaterials show significant promise as contrast agent materials for magnetic resonance imaging (MRI). We have developed a new highly efficient one-step synthesis of magnetically-functionalised hollow carbon nanofibres, where the size of magnetite nanoparticles formed on carbon nanofibres is controlled by the mass ratio of the magnetite precursor (iron acetylacetonate) to nanofibres which act as templates and supports for the nucleation and growth of the magnetic nanoparticles. The physicochemical and magnetic properties of the new hybrid nanomaterials are evaluated and optimised with the aim to ultimately produce an efficient “smart” MRI theranostic agent – a material that allows the combined diagnosis (with MRI), treatment (with magnetic targeting) and follow-up of a disease (with MRI) – currently in high demand for various clinical applications. We have shown that our magnetite-nanofibre materials are effectively solubilised in water resulting in a stable suspension that has been employed as a “negative” contrast agent with an excellent transverse relaxivity (R2) of (268 ± 13) mM/s. Moreover, their attractive magnetic properties make them especially suited for magnetic tracking and thus of particular interest for personalised nanomedicine.

The authors are grateful to Ministry of Education and Science of the Russian Federation. Grant number RFMEFI60715X0132.



Longitudinal r1 and transverse r2 relaxivity.png

Cytotoxicity of ICD-85 NPs on Human Cervical Carcinoma HeLa Cells through Caspase-8 Mediated Pathway

Friday, 30th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 148

Prof. Abbas Zare Mirakabadi [1]

1. *Razi Vaccine and Serum Research Institute*

The biological application of nanoparticles (NPs) is a rapidly developing area of nanotechnology that raises new possibilities in the treatment of human cancers. The cytotoxicity of ICD-85 was evaluated by MTT and LDH assays. The apoptotic effect of free ICD-85 and ICD-85 NPs on HeLa cells was assessed using caspase-8 colorimetric assay. The MTT assay showed that ICD-85 NPs could enhance the in-vitro cytotoxicity against HeLa cells compared to the free ICD-85. The IC50 value at 72 h was reduced from 25 ± 2.9 µg/mL for free ICD-85 to 15.5 ± 2.4 µg/mL for ICD-85 NPs. However, LDH assay demonstrated that ICD-85 has dose-dependent cytotoxicity on HeLa cells while ICD-85 NPs exhibited weaker cytotoxicity on same cells. The results also indicate that ICD-85-induced apoptosis on HeLa cells is associated with the activation of caspase-8. Moreover, caspase-8 assay analysis demonstrated that the ICD- 85 NPs induced a higher apoptotic rate in HeLa cells compared to free ICD-85. Our results demonstrated that the encapsulation of ICD-85 enhances its anti-proliferative effects. Taken together, these results suggest that the delivery of ICD-85 in nanoparticles may be a promising approach for low toxicity and high affectivity in the treatment of cancer

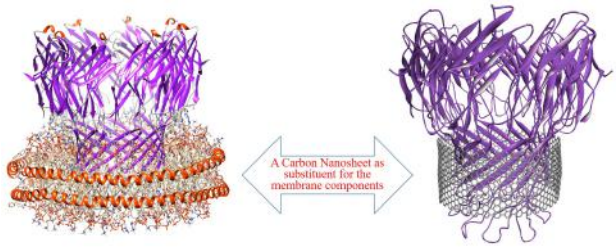
Will a Carbon Nanosheet serve as a replacement to membrane components in a Nanodisc?

Friday, 30th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 206

Dr. Suresh Vepuri [1], ***Prof. Mahmoud Soliman*** [1], ***Prof. Thirumala Govender*** [1]

1. *Discipline of Pharmaceutical Sciences, College of Helath Sciences, University of KwaZulu-Natal, Westville, Durban, South Africa*

Nanodisc is a nano size architecture of an intrinsic biological membrane protein. This in vitro nano biological science process is rapidly evolved into a technology that produce solubilized membrane proteins in their native environment for biomedical and drug discovery applications. The organization of nanodiscs involve a protein enclosed phospholipid bilayer system held together by membrane scaffold proteins (MSPs). MSPs are condensed forms of apolipoprotein (apo) A-I which wrap around a patch of a lipid bilayer to form a disc-like particle or nanodisc. MSPs basically act as a ribbon/sheet to tie the lipid assembly, and simultaneously provide aqueous solubility for the system. Therefore, several engineered MSPs are designed and evaluated by reconstitution. Typically, the MSPs are produced by using recombinant technology and protein purification. To simplify the process, we are interested in designing simple organic structures as substitute for the total membrane components that include both MSPs and lipid layer. As an initiative we attempted a molecular modelling study to assess the propensity of a carbon nanosheet as supporting membrane structure for the protein to form a nanodisc. In this process, we identified a bacterial outer membrane protein porin as example structure to model the carbon nanosheet variant of its nanodisc (Fig 1.). A molecular dynamics study was performed to compare the binding interactions and conformational stability between the native nanodisc model of porin and the nanosheet variant. The nanosheet protein variant was found to be stable and offered a strong hydrophobic interaction network for the protein assembly. Our preliminary study results are interesting and strongly support our idea of nanosheet frame work as substitute for biological membrane system in current nanodisc technology. We anticipate that the carbon nanosheet with its native hydrophobic design and proven flexibility in surface chemical modification, shall be a good alternate for MSPs and could bring new horizons in nanodisc technology.
Acknowledgements: NRF-KIC Travel grant, UKZN-College of Health Sciences, UKZN-Nanotechnology Platform and CHPC Capetwon, South Africa.



Graphene varient of nanodisc.jpg

Accumulation of Doxorubicin Conjugates with Dendritic Polymers and Vector Protein in Normal and Tumor Cells in vitro

Friday, 30th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 223

Prof. Irina Zamulaeva [1], ***Ms. Olga Matchuk*** [1], ***Dr. Nikita Yabbarov*** [1], ***Ms. Elena Nikolskaja*** [1]

1. *National Medical Research Radiological Centre of the Ministry of Health of the Russian Federation*

Introduction

Anticancer agents available today on the market cannot guarantee satisfactory results. There are a number of factors that reduce the efficacy of the drugs. On the one hand, one of the ways to overcome these problems is using increased doses of a drug or application of high toxic agents, which often leads to serious side effects and raises the risk of multidrug resistance occurrence. But, on the other hand, one of the most effective ways to enhance the effectiveness of anticancer drugs is development of new methods for target delivery of chemotherapeutic agents into tumor cells. The aim of this study was a comparative analysis of accumulation of dendritic polymers conjugated with anticancer drug and vector protein in breast cancer and normal cells in vitro. In addition, removal of these drugs from cancer cells was also studied.

Methods

Polyamidoamine dendrimers of the second generation (G2) were used. Accumulation of doxorubicin (Dox), its conjugates with G2-dendrimers (G2-Dox) and recombinant third domain of alpha-fetoprotein as vector protein (3D-G2-Dox) in normal and cancer cells was studied by flow cytometry and laser confocal microscopy. The study objects were cells of peripheral blood mononuclear fraction of healthy donors and cells of breast adenocarcinoma lines MCF-7 and MCF-7/MDR1 differing in chemosensitivity.

Results

G2-Dox and 3D-G2-Dox accumulated in cancer cells of the both lines better than free Dox ($p<0.05$). However, removal of these drugs out of MCF-7 and MCF-7/MDR1 cells was significantly different: in the latter case free Dox was completely excluded from the cells in 24 hours, while Dox accumulating in composition with dendrimers still remained in the cells. It is important that 3D-G2-Dox (unlike G2-Dox) accumulated in normal cells worse than free Dox ($p<0.01$).

Discussion

The results indicate that using of 3D-G2-Dox is the most promising course because it accumulates in cancer cells better but in normal cells worse than free Dox. Furthermore, it can be assumed that applying of 3D-G2-Dox would be especially useful in cases of multi-drug resistance associated with the high expression of P-glycoprotein.

This work was supported by grant of Russian Scientific Foundation # 15-15-10013.

Cytotoxic Effects of Ionizing Radiation and Doxorubicin Conjugates with Dendritic Polymer and Vector Protein on Breast Cancer Cells in vitro

Friday, 30th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 228

Ms. Olga Matchuk [1], ***Ms. Kristina Churyukina*** [1], ***Dr. Nikita Yabbarov*** [1], ***Ms. Elena Nikolskaja*** [1], ***Prof. Irina Zamulaeva*** [1]

1. *National Medical Research Radiological Centre of the Ministry of Health of the Russian Federation*

Introduction

Dendrimers are perspective nanocarriers for anticancer drugs, but unfortunately their conjugates and complexes with drugs lack a targeted action that limits their application. Conjugation of dendrimers with ligands for specific receptors on the tumor cells surface seems promising approach for increasing therapeutic efficiency and decreasing systemic toxicity of anticancer agents. The aim of the study was to evaluate cytotoxic effects of dendrimers conjugated with doxorubicin (Dox) and alpha-fetoprotein recombinant third domain (3D) as vector protein on cancer cells after single and combined application in vitro. Combined effects of ionizing radiation and dendrimers are of great interest because radiation therapy is well known to be used in the treatment of cancer patients in about half of all cases.

Methods

Polyamideamine dendrimers of the second generation (G2) were used after conjugation with 3D and Dox. The cytotoxic effects and intracellular distribution of Dox were studied by MTT-test, light and laser confocal microscopy 24 hours after ionizing radiation exposure of breast adenocarcinoma cells (MCF-7 and MCF-7/MDR1 lines) following by incubation with free Dox, G2-dendrimers loaded with Dox (G2-Dox), or conjugates of G2-dendrimers with 3D and Dox (3D-G2-Dox) during 2 hours.

Results

G2-Dox and 3D-G2-Dox significantly decreased number of MCF-7 cells by 10% and number of MCF-7/MDR1 cells by 25-30% as compared to control ($p<0,05$). Thus, free Dox exerted more pronounced cytotoxic effect on MCF-7 cells and the same effect on chemoresistant MCF-7/MDR1 cells as compared to the conjugates. For MCF-7 line using both conjugates in combination to irradiation was not effective because the conjugates did not increase the cytotoxic effects of radiation exposure. For MCF-7/MDR1 line subadditive effects were shown after the combined treatment with each drug and radiation. The highest synergy factor (0.91) was found for 3D-G2-Dox.

Discussion

In terms of overall cytotoxic effects on stable tumor cell lines, using the studied conjugates is justified only in combination with irradiation and only in the case of high expression of P-glycoprotein, which causes the multidrug resistance of cancer cell line MCF-7/MDR1.

This work was supported by grant of Russian Scientific Foundation # 15-15-10013.

Mutagenicity testing in non-transformed and transformed human breast cell lines after exposure to silver nanoparticles in combination with aluminum chloride, butylparaben, or di-n-butylphthalate

Friday, 30th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 292

Dr. Maciej Stępnik [1], ***Ms. Katarzyna Domeradzka-Gajda*** [1], ***Dr. Joanna Roszak*** [1], ***Dr. Anna Kozajda*** [1], ***Ms. Anna Smok-Pieniążek*** [1], ***Prof. Jarosław Grobelny*** [2], ***Dr. Emilia Tomaszewska*** [2], ***Prof. Grzegorz Celichowski*** [2], ***Dr. Małgorzata Cieślak*** [3], ***Dr. Dorota Puchowicz*** [3]

1. *Department of Toxicology and Carcinogenesis, Nofer Institute of Occupational Medicine, Łódź,* ***2.*** *Department of Materials Technology and Chemistry, University of Łódź,* ***3.*** *Scientific Department of Unconventional Technologies and Textiles, Textile Research Institute, Łódź*

Introduction

Application of silver nanoparticles (AgNP) in cosmetic products is growing rapidly, however very little is known about their biological interactions with other cosmetic ingredients. Considering in vitro and in vivo studies demonstrating accumulation of AgNP in tissues and DNA damage in mammalian cells following exposure to AgNP, the safety issue of using different cosmetic formulations containing AgNP becomes an urgent issue. The aim of this study was to assess potential mutagenic effects of AgNP in combination with other ingredients/impurities of cosmetic products such as: aluminum chloride (AlCl3), butylparaben (BPB) and dibutylphthalate (DBPH).

Methods

AgNP were characterized using the UV-Vis spectroscopy, dynamic light scattering (DLS) and scanning transmission electron microscopy (STEM). The mean size of metallic core of two citrate stabilized AgNP was 15 ± 3 nm and 45 ± 10 nm. The hydrodynamic size of the particles was 19 ± 4 nm and 58 ± 10 nm. Mutagenicity of the AgNP in combination with AlCl3, BPB, or DBPH in three human cell lines, i.e. MCF-7, MDA-MB-231 (both cell types representing breast adenocarcinoma, estrogen receptor(ER)-positive and ER-negative, respectively) and MCF-10A (non-transformed cells), was measured using the in vitro micronucleus test. For comparison, an effect of silver nitrate was also assessed.

Results

Considerable differences in cytotoxicity of both AgNP on normal and cancer cell lines were seen. For example, the highest acceptable concentration (% cytostasis <55%) of AgNP15 used for a 24 h exposure of MCF-10A cells was 47.1 μg/ml, while for MCF-7 and MDA-MB-321 it was 1 μg/ml or 1.2 μg/ml, respectively. The nanoparticles did not show any mutagenic effect neither in combination with the AlCl3 (≤500 μM), BPB (≤200 μM) nor DBPH (≤35 μM). Silver nitrate did not show any mutagenicity at up to acceptable concentrations of 1.5 μg/ml (MCF-10A) or 1 μg/ml (MCF-7 and MDA-MB-321).

Discussion

The results indicate that the AgNP do not show mutagenic activity in the breast cells lines and that selected

cosmetic additives/impurities do not modulate the mutagenic/cytotoxic potential of AgNP.

Funded by the Polish National Science Centre (grant 2012/07/B/NZ7/04197). In this study the apparatus purchased in POIG.01.03.01-00-004/08 project were used.

Electrospun PCL nanofibers loaded with carvacrol for wound dressing applications

Friday, 30th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 294

Mr. Enrique Gamez Herrera [1], ***Dr. Silvia Irusta*** [1], ***Dr. Manuel Arruebo*** [1]

1. *University of Zaragoza*

Introduction

In chronic wounds, an appropriate protection is necessary for a correct healing and actual passive wound dressings are not suitable in these cases. For acute or chronic wounds properties that reduce healing times and associated pain at the same time that maintain humidity, pH and optimum temperature are required. An important property of dressing materials is the formation of an effective barrier against the entry of pathogens.

Essential oils (of thyme, cinnamon, sage, etc.) have antimicrobial properties and some also have demonstrated anti-inflammatory, analgesic and healing properties. Besides they present lower cytotoxicity against eukaryotic cells, and a reduced tendency to promote bacterial resistance than synthetic antimicrobials (e.g. antibiotics).

Electrospinning is a fast and easy method to obtain nanofibers whit high surface area, high surface to volume ratio and good mechanical properties. Biocompatible synthetic and natural polymers as poly-caprolactone (PCL) and chitosan can be used. Furthermore, by this process nanofibers can be loaded with the mentioned natural compounds.

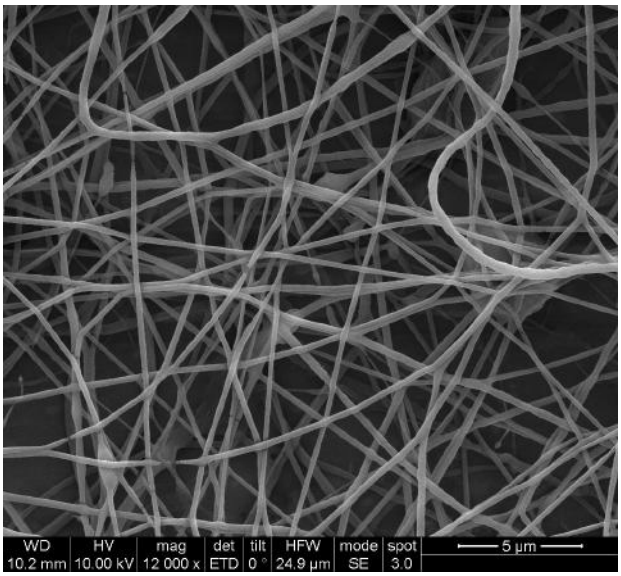
In this work a fast and easy method to produce PCL nanowebs loaded with carvacrol, a natural active compound that have antimicrobial properties, is reported. This compound was selected because minimal bactericidal concentration (MBC) values are below 0.2 mg/mL and 0.4 mg/mL against S. Aureus and E. Coli respectively.

Methods

For the electrospinning process a 10% w/w PCL solution in DCM:DMF 1:1 has been prepared. Then, the appropriate amount of carvacrol has been added to the solution to obtain a 20% w/w in the fibers. Feeding flowrate was 1 mL/h, nozzle-to-plate-distance was 18 cm and voltage used was the necessary to stabilize the Taylors's cone. Drug loading achieved has been determined by GC-MS and the obtained fibers have been characterized by SEM and FTIR.

Results and conclusions

The average diameters of the obtained PCL nanofibers were: 240±40 nm. FTIR spectra show bands related to the natural compounds beside those of PCL corroborating the formation of the composite. Essential oil loading measured by GC-MS was 11.15±0.97% w/w.



Pcl10car20.jpg

Evaluation of Epigenetic Changes in Repetitive Sequences of Human DNA Inducted by Nanoparticles: A Pilot Study for Nanoparticle-Epigenomics Interaction

Friday, 30th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 305

Dr. Amornpun Sereemaspun [1], ***Ms. Siwaporn Nilyai*** [2]

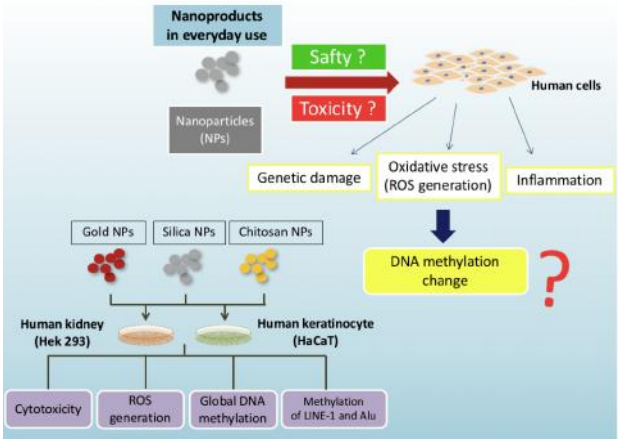
1. *Department of Anatomy, Faculty of Medicine, Chulalongkorn University,* ***2.*** *Nanobiomedicine Laboratory, Faculty of Medicine, Chulalongkorn University*

BackgroundEngineered nanoparticles (ENPs) are one of the most nanomaterials that wildly used in various fields including biomedical applications. However, the adverse effects of ENPs on health risk still need to be concerned. ENPs have been reported to be matter that cause cellular damage through either direct or indirect, cellular oxidative stress is one of the most nanotoxicity have been found. Reactive oxygen species (ROS) is cause of cellular oxidative stress that leads to intracellular macro molecules damages and may impact on DNA methylation changes. In this study we aim to investigate the effect of ROS induced by ENPs on DNA methylation that is one important of epigenetic mechanisms. Human embryonic kidney (Hek 293) and human keratinocyte (HaCaT) cells were used as model to expose with three different types of ENPs, AuNPs, SiNPs and CSNPs.

MethodsWe evaluated cytotoxicity of cells by measuring viability, morphology and ROS levels. Global DNA methylation levels were measured by 5-methylcytosine immunocytochemistry staining, and we also investigated DNA methylation levels of retrotransposable elements, LINE-1 and Alu by using combined bisulfite restriction analysis technique (COBRA).

ResultsWe found ROS level was increased in SiNPs exposed HaCaT cells only. DNA hypomethylation of global and Alu elements was showed in cells were exposed with SiNPs and CSNPs in HaCaT cells only. LINE-1 did not change in both of Hek 293 and HaCaT cells. Furthermore, the inversion of Alu DNA methylation level in HaCaT cells exposed with SiNPs and CSNPs was found in pretreated with antioxidant.

ConclusionOur study demonstrated the new insight that DNA methylation of Alu elements represents the global DNA methylation of cell exposed with ENPs. In this study, the alteration of DNA methylation level in ENPs exposed cells is ROS independent and specific to cell types.



Amornpun abstract.png

Niosome nanoparticles loaded with essential oils for wound dressing applications

Friday, 30th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 328

Mrs. Sara García Salinas [1], ***Mrs. Hellen Elizondo*** [1], ***Dr. Víctor Sebastián*** [1], ***Dr. Manuel Arruebo*** [1], ***Dr. Silvia Irusta*** [1], ***Dr. Gracia Mendoza*** [1]

1. *University of Zaragoza*

Introduction: In nanomedicine the improvement of nanoparticulated systems used in the treatment of different pathologies through local, targeted or controlled release has played a crucial role. Niosomes are one of these nanoparticulated systems, vesicles made by self-assembly of non-ionic surfactants, cholesterol and stabilizers such as dicetyl phosphate (DCP). On the other hand, microorganism resistance is becoming one of the main challenges in medicine research. That is why nowadays plants that were believed to have medicinal properties are studied by its bactericidal, anti-inflammatory and regenerative ability in wound healing, among others. Combination of those materials could be used in topical wound care in order to obtain more targeted and efficient approaches.

Methods: The Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC) of essential oils (EO) reported as bactericidal in Escherichia coli and Staphylococcus aureus were evaluated before being encapsulated in niosomes. EO tested were Carvacrol, Eugenol, Cinnamaldehyde, Tymol, Squalene, Tyrosol and Rosmarinic acid. Inhibitory and bactericidal effects were analyzed by the dilution method using agar broth cultures by contacting different concentrations of EO with bacteria for 24 hours.

Results: The antibacterial effects of EO tested are shown in Table I. It can be seen that Carvacrol, Cinnamaldehyde and Tymol showed higher antibacterial efficiency in both E. coli and S. aureus, being MIC and MBC between 0.1 and 0.5 mg/mL. On the other hand, Eugenol displayed higher bactericidal effects in E. coli than in S. aureus. In this project, we have also carried out the niosome synthesis through a methodology based in microfluidics, using an interdigitated micromixer. Vesicle sizes are 238 ± 41 nm and their Z potential is -33 ± 3mV.

Discussion: In spite of all the efforts that have been made towards the development of artificial wound dressings, none of the currently available options combine all the requirements necessary for quick and optimal cutaneous regeneration. Further studies will be carried out to test the regenerative and anti-inflammatory ability of these EO and to encapsulate them into niosomes, so that the combination may make optimum smart bioactive dressings with a controlled release of the natural substrates.

	Escherichia coli		*Staphylococus aureus*	
	MIC (mg/mL)	MBC (mg/mL)	MIC (mg/mL)	MBC (mg/mL)
Carvacrol	0.2	0.4	0.1	0.2
Eugenol	0.4	0.5	1.2	1.3
Cinnamaldehyde	0.2	0.3	0.4	0.5
Tymol	0.1	0.3	0.1	0.2
Squalene	> 4.0	> 4.0	> 4.0	> 4.0
Tyrosol	> 4.0	> 4.0	> 2.0	> 2.0
Rosmarinic acid	> 4.0	> 4.0	> 4.0	> 4.0

Table i.png

Simple and Efficient Approach for siRNA Encapsulation into PCLs by Freeze-thawing Method

Friday, 30th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 337

Ms. Ayaka Okamoto [1], ***Dr. Hiroyuki Koide*** [1], ***Mr. Hiroki Tsuchida*** [1], ***Dr. Hidenori Ando*** [1], ***Ms. Saki Ariizumi*** [1], ***Ms. Chiaki Kiyokawa*** [1], ***Mr. Masahiro Hashimoto*** [1], ***Dr. Tomohiro Asai*** [1], ***Dr. Takehisa Dewa*** [2], ***Prof. Naoto Oku*** [1]

1. *University of Shizuoka School of Pharmaceutical Sciences,* ***2.*** *Graduate School of Engineering, Nagoya Institute of Technology*

[Introduction] Small interfering RNA (siRNA) has been widely studied for the treatment of intractable diseases, such as cancer. Since naked siRNA is unstable in plasma and has low transfection efficiency, the siRNA delivery system is indispensable for the establishment of systemic siRNA therapy. In the present study, we report an effective and easy-to-use approach for siRNA encapsulation into liposomes by freeze-thawing. We found that freeze-thawing of single-layer polycation liposomes (PCLs)/siRNA complex forms multi-layer structures, suggesting siRNA was effectively packaged between the lipid layers of multi-layer PCLs.
[Method] Dicetyl phosphate-diethylenetriamine conjugate-based PCLs were incubated with siRNA to form PCLs/siRNA complexes (conventional lipoplex). The lipoplex was frozen in liquid nitrogen and thawed in a water bath with vortexing to prepare freeze-thawed lipoplex. Structure of the freeze-thawed lipoplex was observed by transmission electron microscopy. To confirm stability of the siRNA, conventional or freeze-thawed lipoplexes were incubated with fetal bovine serum for 72 h, and then undegraded siRNA was extracted and detected by electrophoretic assay. Gene silencing effect of the freeze-thawing lipoplex was evaluated by reporter gene assay using B16F10 murine melanoma cells transduced with firefly luciferase gene. In order to determine the biodistribution of siRNA in the freeze-thawed lipoplexes, alexa750-modified siRNA (alexa-siRNA) formulated in PEGylated freeze-thawed lipoplex was administrated intravenously to tumor-bearing mice. Then, accumulation of the alexa-siRNA in each organ and tumor was detected by in vivo imaging system.
[Result and Discussion] The transmission electron microscopic observation showed that the freeze-thawed lipoplex formed a multi-layer structure after the freeze-thawing of single-layered PCLs/siRNA complex. The siRNA formulated in freeze-thawed lipoplexes was not degraded even after incubation with 90% fetal bovine serum for 72 h while siRNA formulated in the conventional lipoplexes was degraded. These results indicate that siRNA was packed between the lipid layers and prevented from exposure to nucleases. The freeze-thawed lipoplex showed significantly higher gene-silencing efficacy compared with the conventional ones. Additionally, more amount of alexa-siRNA formulated in PEGylated freeze-thawed lipoplexes showed long-term blood circulation and accumulated in tumor compared with the PEGylated conventional lipoplexes. Our results provide an effective strategy for systemic siRNA delivery with a quite simple procedure.

Adsorption of Bilirubin by Chitosan Coated Activated Carbon Prepared from Date Pits

Friday, 30th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 345

Ms. Ameera Seyedzadeh [1], ***Ms. Asil Mwafy*** [1]

1. *United Arab Emirates University*

The aim of this process was to prepare activated carbon (AC) from date pit powder and observe its adsorption efficiency of bilirubin toxin. Adsorption plays a vital role in the removal of toxins from the blood stream of patients with liver failure. It has been acknowledged from preceding literature that AC has a high capacity to adsorb albumin-bound toxins, which is why it is expended for this purpose. In order to increase the adsorption of bilirubin, an increase in the surface area of the AC was necessary. This increase was achieved through pyrolysis. Furthermore, to increase the capacity of absorbance, the AC was coated with chitosan gel, which contains several groups on its chains that act as interaction sites. Results indicated that the presence of the AC lead to a dramatic cut in bilirubin and the more the AC added to a sample, the faster the rate of adsorption as well as a higher capacity of adsorption allowable.

Using MEMS Resonant Mass Measurement to Characterize Mass, Density and Count of Nano-scale Particles

Friday, 30th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 353

Dr. Hanna Jankevics Jones [1], ***Mrs. Rachel Bott*** [1], ***Mr. Stephen Carrington*** [1], ***Dr. Matthew Barea*** [1]

1. *Malvern Instruments*

Introduction

Using the innovative method of resonant mass measurement (RMM), we present data showing how mass, density and concentration from particle count–for particles ranging from nanometers up to micrometers can be determined. The technology utilizes a suspended MEMS microchannel resonator, through which individual particles transit across the resonator altering the resonant frequency, which is then detected using an optical-based method. Changes in frequency observed when a particle enters the microchannel resonator are proportional to the buoyant mass of the particle, and can be translated into mass, size or surface area[1]. RMM therefore allows particle concentration to be determined as well as particle mass in a single measurement.

Furthermore, RMM enables differentiation of mixtures of particles types based on density differences. For example, silicone oil droplets found in therapeutic pre-filled syringes can be discriminated from protein aggregates, thus enabling accurate quantification of both silicon oil content and protein aggregate concentration[2]. Folzer et al have also shown differences in aggregate densities for protein particles stemming from different proteins such as BSA and IgG[3]. RMM also has the capability of resolving mass differences between uncoated and coated nano-scale particles, as shown by the binding of proteins to latex beads[4].

Methods & Results

Exemplifying the technique, we show how size and concentration of among other gold nanoparticles in suspension can be determined, as well as particle density determination of a range of materials including polystyrene latex, mesoporous silica and metal nanoparticles.

Discussion

Mass is an inherent attribute of all particles, and exploiting this as a detection technique offers a more universal method for characterizing nano-scale particles in a wide variety of dispersants. The ability to measure the particle density is critical for characterization of nano particles, to understand and optimize the production of nano particles for applications such as for example drug delivery, diagnostics or imaging.

REFERENCES

[1] T.P. Burg,M. Godin,S.M. Knudsen,W. Shen,G. Carlson,J.S. Foster,K. Babcock and S. Manalis,Nature 446,1066-1069,2007.

[2] A.R. Patel,D. Lau,and J. Liu, Anal. Chem,84,6833-6840,2012.

[3] E. Folzer,T. Khan,R. Schmidt,C. Finkler,J. Huwyler,H-C. Mahler,and A.V. Koulov,J.Pharm.Sci.,1-7,2015.

[4] M.R. Nejadnik and Wim Jiskoot,J.Pharm.Sci,1-7,2014.

Polydopamine, a potential mucopenetrative nanomaterial capable of multimodal therapy for bladder cancer

Friday, 30th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 384

Ms. Barbara Poinard [1], ***Mr. Samuel Neo*** [2], ***Ms. Angeline Tan*** [2], ***Mr. Roy Tan*** [2], ***Prof. Koon Gee Neoh*** [3], ***Dr. James Kah*** [3]

1. *NUS (National University of Singapore) Graduate School of Science and Engineering,* ***2.*** *Ngee Ann polytechnic,* ***3.*** *NUS (National University of Singapore)*

The high rate of treatment failure for superficial Bladder cancer is partly due to the short residence time of drugs and low uptake, both of which can be explained by urine voiding as well as the presence of a thin mucus layer on the surface of the urothelium. Hence, the development of novel therapeutics that would improve drug retention for patients with bladder cancer is significant.

The present study aims to exploit the many properties of polydopamine (PDA) to design a biodegradable muco penetrative polymeric nanocarrier of Ce6, a photosensitizer drug. PDA is hydrophilic and slightly negatively charged which minimizes hydrophobic interactions and electrostatic interactions with the negatively charged mucus mesh. Furthermore, it is a natural mimic of melanin, biocompatible, easily synthesized and capable of photothermal properties thus enabling multimodal therapy (photodynamic (PDT) and photothermal (PTT)) to increase the drug retention and hence cell kill efficacy of non-muscle invasive bladder cancer cells.

To date, we have successfully characterized and optimized PDA loaded with Ce6 in vitro and studied many parameters such as their loading capacity, stability in different media and release profiles. It has been demonstrated that PDA could effectively load up to 3 microM of Ce6 with an initial burst release and a saturation at day 7. The therapeutic effect of PDA-Ce6 has also been compared to free Ce6 and PDA alone in bladder cancer cells with a significantly higher cell kill when subjected to the dual PDT-PTT. Furthermore, preliminary studies have been performed probing the interaction between mucin and PDA. Results with the Transwell system seemed to indicate that PDA were capable of permeating through the mucus mesh.

The development of these mucopenetrative particles could possibly also be applied in other kinds of cancer or drug delivery therapy which requires passage through the mucus barrier.

Development of a Long-circulating Liposomal Carrier Coated with Serum Albumin via Ligand

Friday, 30th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 403

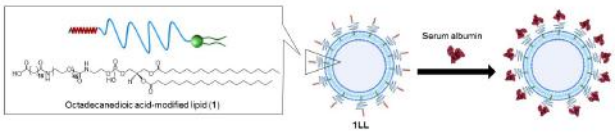
Ms. Hikari Sato [1], ***Mr. Yuta Nakamura*** [1], ***Prof. Akihiro Kishimura*** [2], ***Prof. Takeshi Mori*** [2], ***Prof. Yoshiki Katayama*** [2]

1. *Graduate school of System Life Science, Kyushu University,* ***2.*** *Department of Applied Chemistry, Kyushu University*

Introduction: Polyethylene glycol (PEG)-modified liposome is known as the most succeeded drug carrier because of its long blood half-life. However, it has a problem with its repeated administration because it is rapidly removed from blood after the second administration by produced antibodies against PEG. Here, we focused on serum albumin (SA) for an alternative molecule to provide stealth property to liposome. SA is the most abundant protein in plasma (40 g/L). Since SA can avoid glomerular filtration and degradation in endocytosis, it has a long blood half-life (20 days). Moreover, SA works as an endogenous drug carrier for exogenous drugs and can avoid being caught by immune system. We utilized SA to camouflage the surface of liposome to prolong its blood circulation time. Here, we propose modification of the liposome surface via SA-specific ligands. As such a ligand, we selected octadecanedioic acid, which is reported to bind to albumin. Because of the remaining terminal carboxyl group of octadecanedioic acid after modification of PEG-lipid, insertion of the ligand into hydrophobic liposome's bilayer will be prevented.

Method: Octadecacnedioic acid-modified lipid (1) was synthesized. Then, a liposome containing lipid 1 (1LL) was prepared by a hydration method. To confirm the stability of 1LL in physiological saline, 1LL was incubated in DPBS containing SA from bovine (BSA) at r.t. Change of liposome's size was monitored for a week.

Results and Discussion: In the absence of BSA, 1LL aggregated in DPBS after 2 days. On the other hand, in the presence of BSA, 1LL kept narrow size distribution and dispersed stably for a week. It was speculated that the aggregation of 1LL was caused by hydrophobic interaction between ligands, however, the hydrophobic interaction seems to be suppressed by coating of 1LL with SA. This result indicates that 1LL was successfully coated with SA, contributing significant stabilization of 1LL in physiological saline. 1LL would be applicable to a drug carrier with enhanced blood circulation. Result of blood circulation time of the liposome in mice will be presented.



Iconan.jpg

CNTs recruit different serum protein assortments on the biocorona

Friday, 30th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 412

Mrs. ESPERANZA PADÍN GONZÁLEZ [1], ***Mrs. Nerea Iturrioz*** [1], ***Mrs. Eloisa González-lavado*** [1], ***Ms. Carmen Pesquera*** [2], ***Mr. Fernando González*** [2], ***Ms. Monica L. Fanarraga*** [1]

1. *Universidad de Cantabria-IDIVAL,* ***2.*** *Universidad de Cantabria*

Carbon nanotubes (CNT) have a highly reactive surface that has the intrinsic ability of recruiting surrounding proteins on their surfaces through non-covalent interactions. The CNT biocorona has a chief role in CNT toxicity and bioaccumulation. This protein coating has many effects in vivo providing the nanotube with different biomimetic identities,1,2 dictating the bio-distribution,3 being critical in cellular recognition and CNT biodegradation.4

The formation of the protein corona has been extensively studied in recent years but the underlying biochemical processes taking place on the proteins that bind the nanotube are not completely understood. The interaction of proteins with the CNTs depends on the actual physicochemical properties of the surface of the nanotube,5,6 and also on the structure of the interacting proteins. Most soluble proteins (such as serum albumin) have hydrophobic cores that are only exposed after pseudo-denaturation.7 The exposure of the polypeptide core by partial unfolding of the protein depends on many factors, among others, (i) temperature, (ii) environmental reducing conditions, (iii) time, (iv) solvents, (v) detergents, etc.8 Accordingly, one single protein can display different interactive behaviors with the same nanomaterial depending on these conditions. Here we show how the protein corona of CNTs functionalized with blood serum, recruits diverse assortments of serum proteins on the nanotube surface when exposed to different conditions during functionalization. Our results reveal that the functionalization of the CNT is qualitative and quantitative dependent on all the above environmental conditions.

(1) Nel, A. E., et al., Nat. Mater. 2009, 8 (7), 543–557.

(2) Monopoli, M. P., et al., Nat. Nanotechnol. 2012, 7 (12), 779–786.

(3) Maruyama, K., et al., Biomed Res. Int. 2015, 2015, 793186.

(4)Gea, C., et al., PNAS. 2011, 41, 16968- 16973.

(5)Du, J., et al., JNN. 2011, 11, 10102–10110.

(6)Sopotnik, M., et al., Carbon. 2015, 95, 560-572.

(7)Sengupta, B., et al., RSC Advances. 2015,5, 82395-82402.

(8)Arakawa, T., et al., Adv. Drug Deliver. Rev. 2001, 46, 306-326.

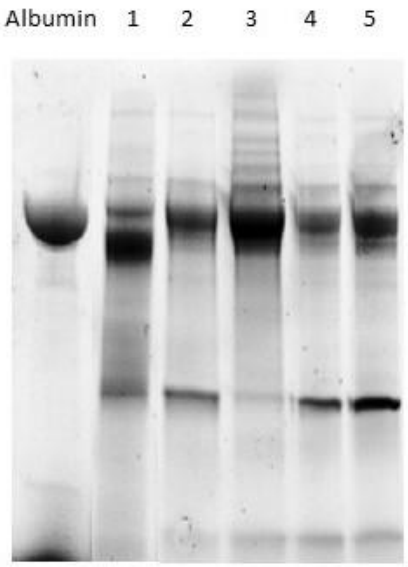


Figure 1. **Functionalization of Nanocyl® NC3100™ MWCNTs with blood serum under different conditions reveals specific biocorona.** SDS-PAGE electrophoresis of CNTs functionalized with serum proteins

Figure1 abstract.jpg

In vitro correction of congenital disorder of glycosylation type Ia (CDG Ia) using PLGA nanoparticles loaded with GDP-Man.

Friday, 30th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 421

Dr. Barbara Bortot [1], ***Dr. Eleonora De Martino*** [2], ***Dr. Alessandra Tesser*** [1], ***Prof. Giovanni Tosi*** [3], ***Prof. Barbara Ruozi*** [3], ***Dr. Giovanni Maria Severini*** [1]

1. *Institute for Maternal and Child Health IRCCS Burlo Garofolo,* ***2.*** *University of Trieste,* ***3.*** *Department of Life Sciences of University of Modena and Reggio Emilia*

Congenital disorder of glycosylation syndrome type Ia (CDG Ia) is an autosomal recessive multisystem disorder with neurological involvement due to mutations into gene sequence that codifies phosphomannomutase 2 (PMM2), enzyme involved in N-glycosylation pathway. Mutated PMM2 leads to a missed conversion of mannose-6P to mannose 1P which results in low concentrations of GDP-MAN in cytosol and little branched oligosaccharide chains. Previous studies have been demonstrating a reduced activity of several lysosomal enzymes such as α-fucosidase, β-hexosaminidase, β-glucuronidase and α–mannosidase in leukocytes and sera of CDG-Ia patients.

We propose an in vitro treatment of CDG fibroblasts using PLGA nanoparticles loaded with guanosine diphosphate D-mannose (GDP-Man), a nucleotide–activated sugar essential to building olygosaccharide chains bypassing glycosylation pathway reaction catalized by PMM2. PLGA-NPS loaded with the commercial guanosine 5'-diphospho-D-mannose sodium salt were prepared by W/O/W (double emulsion) solvent evaporation method. Human fibroblast controls and CDG 1a fibroblasts were obtained from skin biopsies. Specific activities of β-galactosidase, α-mannosidase, β-glucuronidase were measured using 4-methyl-umbelliferyl β-D-galactopyranoside, 4-methyl-umbelliferyl α-d-Mannopyranoside, 4-methyl-umbelliferyl β-d-glucuronide substrates.

In order to determine in vitro hypoglycosylation correction we assayed specific activity of different lysosomal enzymes including β-galactosidase, α-mannosidase and β-glucoronidase from CDG 1a patient's fibroblast cultures. Three fibroblast cultures derived from three CDG patients, called F1, F2 and F3, were incubated with PLGA nanoparticles loaded with 1.5 ug of GDP-Man, corresponding to 2,3 nmol of GDP-Man for three or six hours at 37°C.

The specific activity of lysosomal enzymes like α-mannosidase and β-glucoronidase were assayed at three different times after treatment: 24 hours, 48 hours and 72 hours. As shown in Figure 1, α-mannosidase attains levels of activity close to fibroblast control activity after 48 hours post-incubation in F1, F2 and F3, demonstrating the efficiency of the proposed treatment. To date there is no therapy for CDG-Ia patients, but PLGA nanoparticles loaded with GDP-Man might provide a suitable treatment to bypass PMM2 deficiency and improve protein glycosylation due to their high biodegradability and toxicity profile.

Keywords: Congenital disorder of glycosylation, poly(lactic-co-glycolic acid)
nanoparticles, Guanosine diphosphate and Mannose, phosphomannomutase 2, lysosomal enzyme, specific activity.

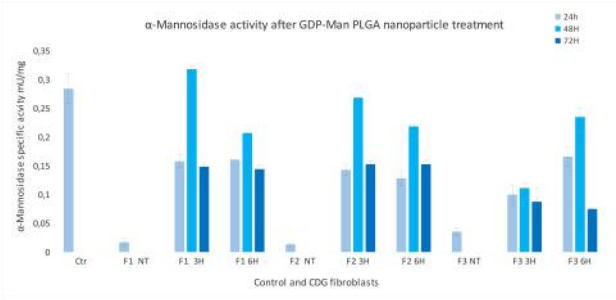


Fig.1: Correction using GDP-Man PLGA nanoparticles of a-mannosidase activity in three cultured fibroblasts obtained from CDG patients. Analysis was repeated three times and the mean ±s.d. is reported.

Gdp-man plga nps correction.jpg

Effect of the nanopartilcle shape on the cancer treatment: the case of pluronic F127 stabilized and doxorubicin loaded magnetite nanocubes and nanospheres.

Friday, 30th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 444

Dr. Timur Nizamov [1], ***Dr. Anastasia Garanina*** [1], ***Prof. Alexander Savchenko*** [1], ***Prof. Alexander Majouga*** [1]

1. *NUST MISiS*

Magnetic nanoparticles are extensively prepared for the potential use in nanomedicine fields: MRI and drug delivery. Magneto-niosomes – magnetic nanoparticles, stabilized by non-ionic surfactants are the ones of the promising agents in such fields. They are examples of theranostic agents combining therapy (drug delivery) and diagnostics (MRI contrast agents) in one agent. The other rarely studied aspect is the effect of nanoparticle shape on toxicity, drug delivery and relaxivity. The following work is dedicated to synthesizing of magneto-niosomes with different core shapes (spherical and cubic), and investigating the effect of nanoparticles shape on relaxivity and toxicity towards PC3 and LNCaP cancer cell cultures.

Nanospheres and nanocubes were synthesized by thermal decomposition of iron oleate in 1-octadecene. Shape control was reached by different stabilizers molar ratio in reaction medium. According to TEM measurements average size of the nanoparticles is 20±5 nm. The synthesized nanoparticles were transferred into water by non-ionic surfactant (Pluronic F127) solution. DLS data demonstrates that nanoparticles average size has risen up to 90±10 nm due to niosomes formation. The hydrophilized nanoparticles were loaded with doxorubicin by addition of its solution into hydrosol and stirring. Doxorubicin excess was taken away by centrifugation and supernatant removing. Spectrophotometry was used to determine the doxorubicin loading (the average drug loading is 13,5% of drug carrier). Loaded niosomes cytotoxicity was measured on PC3 and LNCaP cultures by MTT assay and compared to cytotoxicity of unloaded nanoparticles and free doxorubicin after 48 hour of incubation.

The obtained data demonstrate lower cytotoxicity of doxorubicin loaded nanoparticles compared to free doxorubicin at the same doxorubicin concentrations in cell culture. This happens due to slow doxorubicin releasing from Pluronic shell of magneto-niosomes which play a role of drug carrier. Also nanocubes based niosomes has shown a bit higher cytotoxicity compared to analogous nanospheres based agents. This effect is caused by four times higher uptake of nanocubes versus nanospheres. Both cubic and spherical niosomes demonstrated relatively high T2-relaxivity (240 mM-1*c-1 and 262 mM-1*c-1 respectively).

The authors knowledge financial support from Ministry of Education and Science of the Russian Federation (14.607.21.0132, RFMEFI60715X0132).

Atomic Force Acoustic Microscopy for the Characterization of Gold Nanoparticles Embedded into a Polymeric Matrix and its Promissory Application in Nanomedicine

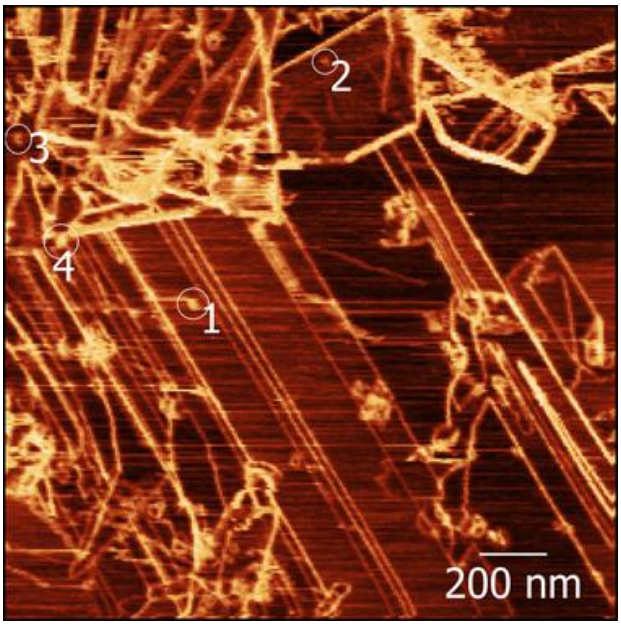
Friday, 30th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 445

Dr. Daniela Geraldo [1], ***Mr. Eduardo Jeraldo*** [1], ***Prof. Ramiro Arratia-perez*** [1]

1. *Universidad Andres Bello*

In this work, we report for the first time a morphological study of gold nanoparticles embedded on a polymeric matrix using Atomic Force Acoustic Microscopy (AFAM). The polymer based-gold nanoparticles were prepared by a green approach using biomass from macroseaweed, as a mixture of reducing agents containing not only water soluble polysaccharides but also phytoactive compounds. It was demonstrated that AFM in the acoustic mode is a powerful tool that allows obtain conclusive information about the morphology of a natural polymer acting not only as a reducing agent but also as stabilizing agent for the gold nanoparticles. The formation of gold nanoparticles was monitored by UV-Vis spectroscopy and Transmission Electron Microscopy (TEM) confirming the presence of monodisperse and isotropic nanostructures. Finally, the hemolytic activity of the gold nanoparticles embedded into the polymeric matrix was tested in order to compare their intrinsic cytotoxicity with those ones prepared using the well-known inorganic reducing agent, sodium citrate (SC). The obtained results strongly suggest that these eco-friendly gold nanoparticles could be used on human cells due its less toxicity which expand their use on nanomedicine.

Acknowledgements: Financial support for this work was provided by the Air Force Office of Scientific Research Project FA9550-12-1-0367 and Proyecto Núcleo UNAB DI-622-14N.



Gold nps embedded into polymer by afam 10x10.jpg

Preparation and characterization of multimeric system of RGD-grafted PMMA-nanoparticles as a targeted drug-delivery system for paclitaxel

Friday, 30th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 484

Ms. Brenda Vianey Gibbens-Bandala [1], ***Dr. Enrique Morales-Avila*** [1], ***Dr. Blanca Eli Ocampo-García*** [2], ***Dr. Guillermina Ferro-Flores*** [2]

1. *Universidad Autónoma del Estado de México,* ***2.*** *Instituto Nacional de Investigaciones Nucleares*

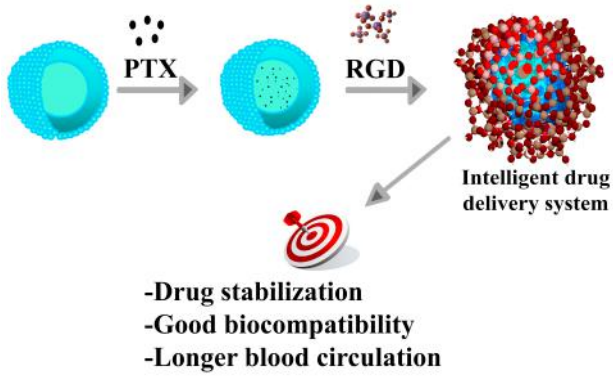
In cancer, therapeutic alternatives include the use of antineoplastic drugs; however, current drugs lack selectivity for only to damaged tissue, which produce severe adverse effects. Improvements in the development of polymer-based nanoparticles as drug-delivery systems, offers new therapeutic opportunities. Among them, ligand-mediated targeting, interacts with specific receptors on target cells, increases selectivity and efficacy and allows controllable drug delivery, increasing therapeutic benefits, while minimizing side effects. Additionally, it offers the possibilities of encapsulating poorly-soluble drugs, protecting therapeutic molecules, and modifying the pharmacokinetic profile of nanoparticle-incorporated drugs.

The aim of this research was to prepare and to characterize poly(methyl methacrylate) (PMMA) nanoparticles grafted with the –Arginine, Glycine, Aspartic Acid (RGD)– peptide sequence as a promising smart drug delivery system for Paclitaxel (PTX), directed to sites with integrin receptor overexpression, such as angiogenesis sites in some solid tumors.

Characterization techniques included FT-IR spectroscopy, particle size by dynamic light scattering (DLS), zeta potential, morphology by transmission electron microscopy (TEM), entrapment efficiency (%EE), drug release profile and cytotoxicity in positive integrin receptor C6 cell line. The vibrational spectroscopy characterization indicated the formation of the PMMA polymer, PMMA-PTX or RGD-PMMA-PTX modified polymer. RGD-PMMA-PTX size distribution was found to be 17.58 ± 7.45 nm with a zeta potential of

-38.73 ± 5.62 mV and of spheroidal form. According to the boxLucas Model, PTX was incorporated into polymeric matrix with an entrapment efficiency maximum of 100% (HPLC). These nanoparticles showed sustained in vitro release with maximum release percentages of 55% and 40% after 21 days at pH 5.3 and 7.4, respectively, a relatively fast release of PTX in the first 5 hours at pH 5.3, but not for pH 7.4 attributable to the diffusion and dissolution of PTX that was entrapped superficially in the polymer matrix. RGD-PMMA-PTX nanoparticles were shown to be more cytotoxic to C6 glioblastoma cells than non-targeted nanoparticles and free PTX.

These results indicate that RGD-PMMA-PTX is suitable for specific PTX delivery in cells that over-express αvβ3 receptors in processes of angiogenesis. In this study, the small amount of PTX required to produce an anti-proliferative effect opens the possibility of reducing side effects.



Preparation of multimeric system of rgd-grafted pmma-ptx nanoparticles.jpg

Effect of Ligand Shell Composition on Biodistribution of Ultrasmall Gold Nanoparticles

Friday, 30th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 506

Dr. Yao Ding [1], ***Dr. Tom Coulter*** [1], ***Ms. Cristina Espinosa Garcia*** [1], ***Dr. Sarah Hale*** [1], ***Mr. Alessandro Pace*** [1], ***Dr. Ketan Patel*** [1], ***Ms. Usoa Aguilera Peral*** [1], ***Mrs. Angela Robinson*** [1], ***Dr. Dan Palmer*** [1], ***Dr. Phil Williams*** [1], ***Dr. Meike Roskamp*** [1]

1. *Midatech Pharma Plc*

Cytotoxic chemotherapy is the standard of care for many types of cancer despite the frequently observed severe side effects. The targeted delivery of chemotherapeutics has great potential to reduce these effects by increasing drug concentrations within the target tissue, thereby reducing the required dose [1]. Targeted delivery can be an extremely useful approach to achieving a therapeutically active concentration whilst ensuring acceptable systemic toxicity [2].

Midatech Pharma Plc uses novel ultrasmall gold nanoparticles (GNPs) with a gold core diameter of 1.6 – 1.8 nm as a functionalizable vehicle for tissue-selective delivery (Figure 1). This platform is highly flexible, allowing incorporation of various components dependent upon the therapeutic application [3]. In the current study we describe GNPs that are coated with thiol-modified carbohydrates and bifunctional polyethylene glycols (PEGs). We investigated the effect of different chemical ligand shell compositions on the biodistribution and pharmacokinetic profile of these particles using an in vivo rat model. By varying the charge and ligand size, we showed that Midatech's novel ultrasmall gold nanoparticles can be customized to vary circulation times and tissue selective accumulation. Also, due to their very small size and carbohydrate based ligand shell these particles are less likely to form a protein corona compared to larger nanoparticle analogues [4], and are therefore especially interesting candidates for active targeting strategies.

In conclusion, Midatech GNPs offer a highly flexible and customizable drug delivery system which may be tailored for a range of cancer types and other diseases.

References

[1] S. M. Sagnella, J. A. McCarroll, M. Kavallaris, Nanomedicine, 10, 1131 (2014).

[2] M. Shilo, M. Motiei, P. Hana, R. Popovtzer, Nanoscale, 6, 2146 (2014).

[3] midatechpharma.com.

[4] K. Zarschler, L. Rocks, N. Licciardello, L. Boselli, E. Polo, K. P. Garcia, L. De Cola, H. Stephan, K. A. Dawson, Nanomedicine, 12 (6), 1663-1701.

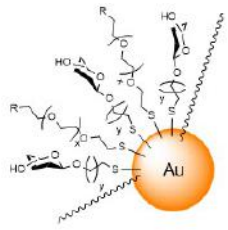


Figure 1. Functionalized gold nanoparticles for drug delivery (R = COOH, NH2).

Generic particle.jpg

Influence of biophysical anomalies on forest tree quality

Friday, 30th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 563

Dr. Aigars Indriksons [1], ***Mr. Maris Eglite*** [1], ***Dr. Imants Liepa*** [1]

1. *Latvia University of Agriculture, Forest faculty*

Biolocation is a method which allows to estimate the biophysical anomalies on the earth surface, underground and air by considering of changes in condition of dowsing rod or other indicator in hands of dowser. The most popular version of biophysical anomalies is connected with flows of underground water. Water has an 81 times higher relative dielectric coefficient as air. However structures of different networks suggest for another nature of radiation which does not connect with water flow.

There is very few research carried out of this theme in Latvia. In 1979 there was the section of Bioelectronics established by the A. Popov's scientific-technically society of Radio engineering, electronics and communication, which in fact worked in Institute of Physics of Latvian Academy of Science.

The research carried out in Liepupe Forestry suggests that the pine (Pinus sylvestris L.) trees growing on biophysical anomalies are more vigorous and those growing on points of intersection are with wider diameter. However, the pine trees measured by City Jelgava gives a converse suggestion – the trees growing on places with biophysical anomalies are thinner and also with lower density.

According to news of Finnish forest scientist V. Altonen, during the 30ties of the 20 century a number of scientists have tried to clarify if there in forest is some influence of biophysical anomalies to the flora. K. Müller (1935; 1936) indicates that trees growing on points of intersection of anomalies are decayed. He has observed that trees growing on the biophysical anomalies are more infected by diseases and more abundant are the agents of diseases – parasitic fungi and bacteria.

Till up to now there are no scientifically secure evidence between any external physical fields and biophysical anomalies obtained. However, if something can not been proven or rejected in scientifically proper way, it does not mean that it is not existing and has not to been explored with increasingly growing technical resources.

Keywords: biophysical anomaly, biolocation methods, tree growth.

Engineered Nanoparticles for Rapid Delivery of Betulinic Acid in MDA MB 231 and HEp-2 Cancer Cell Lines

Friday, 30th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 279

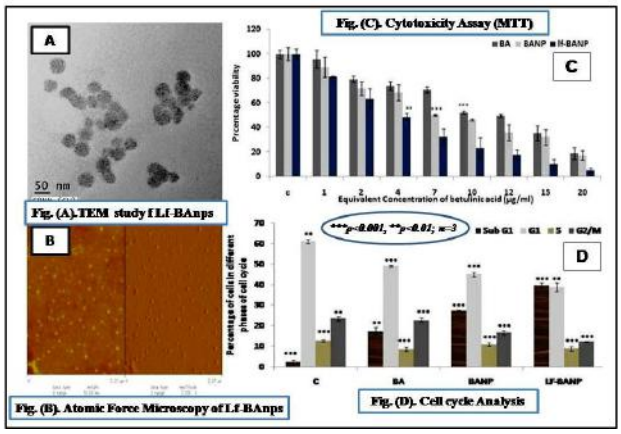
Mr. Asim Halder [1], ***Ms. Pritha Mukherjee*** [1], ***Mrs. Subarna Ghosh*** [1], ***Dr. Urmi Chatterji*** [1], ***Prof. Arup Mukherjee*** [1]

1. *University of Calcutta*

Introduction: A range of triterpenoids have gained attention in cancer chemotherapy due to promising pre-clinical results. Triterpenoids are however constrained in therapeutics for both the solubility and permeability limitations. Betulinic acid (BA) is a pentacyclic triterpenoid available in Betula alba (white birch) bark. BA is enlisted under the RAID (Rapid Access to Intervention Development) program of NCI, USA. The compound is a strong inhibitor of topoisomerase and induces caspase activation and mitrochondrial membrane damage. BA is widely available and economic but suffers seriously in bio-pharmaceutics. Smart nanoparticle drug delivery devices were therefore conceived as one solution for BA delivery. Lactoferrin was used for nanoparticle ligand tethering for successful particle propagation up to the cancer cell nucleus.

Methods: PLGA nanoparticles loaded with BA (BAnps) were prepared in modified solvent diffusion method using PLF 127 as a stabilizer. Lactoferrin conjugation on BAnps (Lf-BAnps) was achieved in EDC/NHS coupling reactions. Nanoparticles were characterized in DLS, AFM, TEM, XRD, FTIR and SDS-PAGE. In-vitro anticancer efficacy was evaluated in two metastatic cancer cell lines: MDA-MB-231 and Hep-2.

Results and Discussion: The average particle size of Lf-BAnps in DLS was 120 nm. The entrapment efficiency of BA (75.42%) and non-fickian release pattern were determined by RP HPLC. The FT-IR and SDS-PAGE analysis confirmed lactoferrin interactions with BAnps. Lf-BAnps showed significantly strong antiproliferative and cytotoxic effects on both the cancer cell lines (IC50 4.0 µg/ml). Time-dependent uptake and quantitative count in flow-cytometry showed early time-point intracellular entry of Lf-BAnps. Specific cellular localization was confirmed by confocal microscopy. Increased sub-G1 population and decreased G2/M population indicating the apoptotic potential and cellular arrest of Lf-BAnps on cancer cells. Rapid localization and lactoferrin bio-functionalization in nanoscale has proved as one successful strategy in chemotherapeutic drug targeting.



Nanoparticles characterization and anticancer evaluation study.jpg

Hybrid biocompatible silica nanoparticles as theranostic agent

Friday, 30th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 122

Mr. Ritu Raj [1]***, Ms. Carina Crucho*** [2]***, Prof. José Paulo Sequeira Farinha*** [2]***, Prof. Carlos Baleizão*** [2]

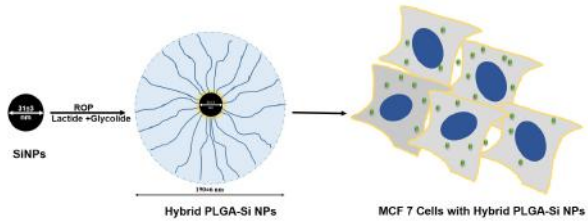
1. *CQFM – Centro de Química-Física Molecular and IN – Institute of Nanoscience and Nanotechnology, Instituto Superior Técnico, Lisboa, Portugal, Department of Life Science, National Institute of Technology, Rourkela, Odisha, India,* ***2.*** *CQFM – Centro de Química-Física Molecular and IN – Institute of Nanoscience and Nanotechnology, Instituto Superior Técnico, Lisboa, Portugal*

Introduction: Hybrid silica nanoparticles have attracted much attention owing to their potential biomedical applications in diagnostic and precision drug delivery for cancer detection and treatment. The inorganic silica core can be used to transport cargo and impart properties such as fluorescence, whereas the polymeric shell can be used to convey water solubility, biocompatibility, long blood circulation times, and bioconjugation. The goal of our work is to develop “smart” hybrid nanoparticles (NPs) with theranostic (therapeutic + diagnostic) functionalities that carry a fluorescent dye for traceability and imaging, feature a mechanism for release control, and are able to accommodate large drug loads and deliver their cargo to the desired location. Here we focus on the preparation of fluorescent silica nanoparticles (SiNPs) with a poly(lactide-co-glycolide) (PLGA) biocompatible and biodegradable copolymer shell.

Method: We prepared monodisperse fluorescent SiNPs incorporating a perylenediimide (PDI) derivative in the silica network, and from the SiNPs external surface, a dense polymer shell of PLGA was grown by ring-opening polymerization (ROP), with different monomers ratio to modulate the degradation rate of the shell. The biocompatibility of the NPs was tested against MCF-7 human breast carcinoma cells. Cell viability and confocal studies were performed to assess the efficacy of the hybrid NPs.

Result & Discussion: The synthesized SiNPs were spherical and homogenous with an average diameter of 31 nm which was confirmed by Transmission electron microscopy (TEM) and Dynamic Light Scattering (DLS). The increased diameter of the Hybrid NPs due to the formation of a PLGA shell on the surface of the silica core has been established by TEM and DLS. The cell viability (MTT assay) showed a high level of biocompatibility, and confocal images confirmed the time-dependent internalization of hybrid NPs into the MCF7 cells.

Acknowledgments: This work was partially supported by Fundação para a Ciência e a Tecnologia (FCT-Portugal) and COMPETE (FEDER), projects RECI/CTM-POL/0342/2012, UID/NAN/50024/2013, and PTDC/CTM-POL/3698/2014. R.R. thanks, Erasmus Mundus Experts Sustain Ph.D. grant.



Abstract for iconan.jpg

Comparison of the effect of selenium Nano particles and sodium selenite on the serum and hepatic lipid profiles of rats following experimental exposure to cadmium.

Friday, 30th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 472

Dr. Saeid KarimiDehkordi [1], ***Dr. Abdonnaser Mohebbi*** [1], ***Prof. Mohammad Reza Aslani*** [1], ***Dr. Afshin Jafari Dehkordi*** [1], ***Dr. Akram Shahabi*** [1]

1. *Shahrekord University*

Introduction:
Cd exposure alters cellular normal process leading to dysfunction in different organs. Several studies have reported the cardiovascular effects of Cd in human and animal models. Some studies have revealed the lipid metabolism alterations in Cd exposed human and experimental animals. Cd causes modification in the blood lipid profile toward a atherogenic condition.
Selenium is known due to its antioxidant function. The reduction of Se salts in presence of proteins forms Se nanoparticles (Nano-Se) which possess greater biological activity than ion form. It is well known that Se alleviates cadmium toxicity by preventing oxidative damage. However the effect of Se on the lipid metabolism in case of Cd toxicity has not been studied.
The aim of present work was to study the probable effect of Se in two different form (sodium selenite, Na-Se, and Nano-Se) on the lipid metabolism in Cd intoxicated rats.
Methods:
Male rats were divided in 4 different experimental groups. A control group and other rats were orally received Cd (3mg/kg b.w) alone or with red Nano-Se (20-30 nm) or Na-Se (0.1 mg/kg b.w) for 35 days. Serum and liver lipid profile were determined using standard methods.
Results:
Cd exposure increased blood and liver TG, total cholesterol, LDL-C and VLDL-C and decreased HDL-C in affected rats compared to control group. Serum and liver TG concentrations were decreased near control group by receiving both Nano-Se and Na-Se, 29% and 38% respectively. The liver total cholesterol and LDL-C contents were not affected by Na-Se while Nano-Se caused a statistically significant reduction on them. Our data revealed that receiving both forms of Se led to decrease in serum concentrations of total cholesterol and its lipoprotein containers, meanwhile, receiving Nano-Se had a relatively greater effect than Na-Se.
Discussion:
Based on our finding, Nano-Se has stronger ameliorative effects on lipid metabolism disorders in Cd intoxicated rats in comparison to Na-Se. The main mechanism of this effects were not clear. However, It is well known that biologic properties of Se Nano-Se specially antioxidant activity, are dependent on the particle size and smaller particles have greater activity.

Mannose Nanoparticles from Guar Gum: Macrophage Vectorization, Drug Delivery and Leishmanicidal Efficacy against Wild and Drug Resistant Strains

Friday, 30th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 244

Prof. Arup Mukherjee [1], ***Mr. Asim Halder*** [1], ***Mr. Sumanta Kumar Ghosh*** [1], ***Ms. Nandita Ghosh*** [1], ***Dr. Ena Ray Banerjee*** [1]

1. *University of Calcutta*

Introduction: Guar gum (GG) is obtained in sufficient purity from Cyamopsis tetragonoloba seeds. Unlike cellulose, GG presents one β 1-4 mannose chain interposed with α 1-6 galactose substituents. GG hydrates hugely in mannose surfaced coiled assembly formations. We are successful for the first time in extracting mannose nanoparticles (GMn) from GG. GMns observed rapid receptor gated uptake in macrophages. Drug loaded GMns were used against macrophage resident L. donovani strains.

Methods: Temperature controlled GG hydrolysis in sulfuric acid (64%) yielded GMns spheroids, TEM size 48.8 nm, ζ potential -20.4mv. Epichlorohydrin in liquor ammonia reactions on GMn provided cationic modifications and fluorescent rhodamin dye was linked following isothiocyanate chemistry. Quercetin (Qr) was loaded in GMns by porous diffusion in phosphate buffer. The loading efficiency in HPLC was 87% w/w and FT-IR studies confirmed nano-chemistry. Qr loaded GMns efficacy against L. donovini AG 83 strains were studied against axenic and macrophage infested amastigotes.

Results and Discussion: GMns and Qr loaded GMns were taken up rapidly in mouse peritoneal macrophages. Uptake was complete within 60 min and 2 mM mannose but not galactose could inhibit that significantly. Concanavallin A agglutination experiments confirmed GMns 42% surface mannose density. Qr loaded GMns were safe and powerful leishmanicidals against wild and drug resistant strains (Table 1).

Table 1. Antileishmanial efficacy of quercetin loaded GMns against Leishmania donovani.

Table 1. Antileishmanial efficacy of quercetin loaded GMns against *Leishmania donovani* AG 83

	IC_{50} (mean ± SD 3 replicates) µM						
	Axenic amastigote			Amastigote in macrophage			
Drug	WT	SSG resistant	PMM resistant	WT	SSG resistant	PMM resistant	Cytotoxicity Macrophage cells CC_{50} µM
Amphotericin B	0.2±0.05[A]	0.4±0.05[A]	0.35±0.05[A]	0.15±0.05[A]	0.2±0.05[A]	0.18±0.05[A]	14
Sodium stiboghuconate (SSG)	3.6±0.40	130±20	115±16	1.6±0.20	18.1±3	17.3±2.60	27
Paromomycin (PMM)	10±2[A]	380±40[A]	330±30[A]	8±2[A]	125±15[A]	115±13[A]	248
Qr loaded GMn	25±4[A]	30±5[B]	26±6.1[B]	18±5[A]	21±3[B]	24±4.2[B]	$6.4x10^3$
Qr	31.2±2.1[A]	145.5±24[A]	75±10[A]	33.0±6.5[A]	123±16[A]	64.0±9.5[A]	$12x10^3$

WT (Wild Type), SSG (sodium stiboghuconate) and PMM (paromomycin). * P<0.05 highly significant, *P>0.5 significant and * P<1 no significant difference compared with SSG group.

Table 1. antileishmanial efficacy of quercetin loaded gmns against leishmania donovani..png

PAMAM dendrimer curcumin conjugate as a supramolecular polyphenol; a biomimetic approach to improve anticancer properties of curcumin

Friday, 30th September - 13:30 - Poster Session & Sponsors Exhibition - Patio 25 - Poster presentation - Abstract ID: 316

Dr. Vahid Erfani-Moghadam [1], ***Prof. Majid Sadeghizadeh*** [2], ***Dr. Alireza Nomani*** [3]

1. *Department of Biotechnology, Faculty of advanced Medical Technology, Golestan University of Medical sciences, Gorgan, Iran,* ***2.*** *Department of Genetics, Faculty of biological sciences, Tarbiat Modares Univesity of Tehran, Tehran,,* ***3.*** *Department Of Pharmaceutical Biomaterials, School Of Pharmacy, Zanjan University Of Medical Sciences*

Introduction: Nanotechnology researchers try to design novel drugs with new properties, such as enhanced bioavailability, biocompatibility and/or biodegradability. Curcumin is a potent natural anticancer agent, but its efficiency is limited by drawbacks, such as very low solubility, high rate of degradation and low rate of absorption of its hydrophobic molecules in vivo. In this study, we investigated the cell uptake and anticancer effects of curcumin on U87MG brain carcinoma cell line and HFSF-PI3, as a normal human fibroblastic cells, by preparing PAMAM (generation 4) dendrimer curcumin conjugates.

Methods: The curcumin conjugation to the surface of PAMAM dendrimers and its conjugation quantity were evaluated by FTIR and NMR methods. Then, cytotoxicity, cell cycle (by PI) and apoptosis (by PI/Annexin-V-FLUOS methods) of various PAMAM-based curcumin conjugates were investigated using U87MG cells. Also, real-time PCR method was used to evaluate the relative gene expression of anti-apoptotic genes; Xiap, RB1 and P21.

Results: The cytotoxicity studies showed that the IC50 of the free curcumin solution, 20% and 40% curcumin-PAMAM conjugated and PAMAM-curcumin encapsulation, were 48, 0.9, 2.5 and 13.5 μM, respectively, on U87MG cancer cell line. Results of the cell cycle and apoptosis analyses indicated that with the curcumin conjugated or encapsulated to PAMAM dendrimer, sub G1 population and apoptosis were significantly increased compared to that of the free curcumin. The results showed that Xiap and P21 gene expression in U87MG cells significantly decreased.

Discussion: Overall, the obtained results showed that dendrimer-based curcumin formulation induces apoptosis in U87MG cancer cells. Results showed that the application of these nanocarriers significantly increased the cell uptake of curcumin in comparison to the free curcumin. At the same time, by conjugating to the PAMAM surface, curcumin obtains a better water stability and perhaps longer therapeutic effect than PAMAM-encapsulated curcumin. In summery, based on the fact that supramolecular polyphenols have strong anticancer effects compared with small molecules, it seems that the synthesis of such big molecules can be considered as an effective biomimetic approach and as an appropriate drug delivery system for curcumin to U87MG cancer cells and probably as a suitable candidate for in vivo researches.

nanoMIL100: a novel efficient tool to target lung cancer

Friday, 30th September - 16:00 - Targeted drug delivery and Nanocarriers - Nanomedecine for cancer diagnosis & therapy - Amphitheatre 25 - Oral presentation - Abstract ID: 48

Dr. Teresa Simon-Yarza [1], ***Dr. Monica Gimenez-marques*** [1], ***Ms. Rhizlaine Mrimi*** [1], ***Ms. Angelika Mielcarek*** [2], ***Dr. Ruxandra Gref*** [3], ***Dr. Patricia Horcajada*** [1], ***Dr. christian serre*** [1], ***Prof. Patrick Couvreur*** [4]

1. *Institut Lavoisier, UMR8180 CNRS/Université de Versailles St-Quentin en Yvelines,* ***2.*** *Ecole Normale Superieure,* ***3.*** *Institut Galien, UMR8612 CNRS/Université Paris-Sud,* ***4.*** *Institut Galien, UMR8612 CNRS, Université Paris-Sud*

Lung cancer (LC) is a major health problem, accounting for near 27% of all cancer deaths. Each year, more people die of LC than colon, breast, and prostate cancers combined (1). This fact evidences the urgent need to improve current available therapy. To increase LC therapy and to reduce its toxicity we propose the use of MOF-based nanoparticles (NP). In particular, we have developed a formulation that passively targets the lungs without any visible signs of toxicity based on MIL-100(Fe), a mesoporous iron(III) trimesate MOF.

NP were prepared and characterized as described elsewhere (2). To evaluate their in vivo biodistribution and toxicity at early times (up to 24 h), a first study was performed in Sprague-Dawley rats. An important accumulation of the NP was observed in the lungs by measuring the iron concentration by atomic absorption spectroscopy. In accordance with our previous data, animals did not present any signs of toxicity. This result encouraged the evaluation of the impact of the loading of Gemcitabine-monophosphate (GMP), a well-known drug used in LC treatment, into MIL100 NP, on drug organ accumulation.

Tritiated-GMP was encapsulated by the impregnation method as previously described (3). C57BL/6JRj mice were treated iv with 10 mg/kg of the free drug and the equivalent dose of MIL100 encapsulated drug. When administered in NP, GMP concentration in lungs was five-fold higher than free GMP after 15 min and remained three-fold larger after 8h (Figure 1).

Thus, MIL100 NP appear as an efficient tool to target the lungs to treat pulmonary diseases.

1. American Cancer Society statistics (last revised 2015).

2. P. Horcajada et al. Chem. Rev., 2012, 112(2) 1232.

3. V. Rodriguez-Ruis et al. J Drug Target. 2015, 23(7-8), 759.

Figure 1 Drug accumulation (measured as disintegrations per second per mg of organ).

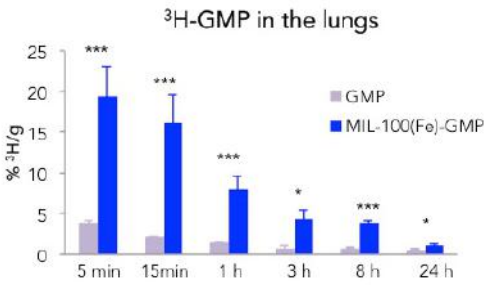

^{3}H-GMP in the lungs
% ^{3}H/g
25
20
15
10
5
0
***
***
***
*
***
*
GMP
MIL-100(Fe)-GMP
5 min
15min
1 h
3 h
8 h
24 h

Figure 1.jpg

Hyperbranched polyglycerol-docetaxel treatments for bladder cancer and the characterization of treated bladder tissue using MALDI-MS imaging

Friday, 30th September - 16:25 - Targeted drug delivery and Nanocarriers - Nanomedecine for cancer diagnosis & therapy - Amphitheatre 25 - Oral presentation - Abstract ID: 257

Dr. David Plackett [1], ***Dr. Clement Mugabe*** [2], ***Ms. Shujun Lin*** [3], ***Dr. Guobin Sun*** [3], ***Dr. Nancy Ford*** [3], ***Dr. Richard Liggins*** [2], ***Prof. Helen Burt*** [1]

1. *Faculty of Pharmaceutical Sciences, University of British Columbia,* ***2.*** *The Centre for Drug Research and Development (CDRD),* ***3.*** *The Centre for High-Throughput Phenogenomics (CHTP), Faculty of Dentistry, University of British Columbia*

Introduction

In North America, bladder cancer is the 4th most common cancer in men and the 9th in women and is reported to have the highest lifetime cost per patient of all cancers. Furthermore, innovations in the treatment of bladder cancer have been slow to develop and there remain significant unmet medical needs. The use of mucoadhesive nanoparticles based on amine-modified hyperbranched polyglycerols (HPGs) as vehicles for delivery of the taxane drug docetaxel (DTX), including drug uptake, efficacy, safety and pharmacokinetics, has been the subject of our recent research. In this context, the aim of the study reported here was to demonstrate that DTX could be visualized and quantified in bladder tissue by means of matrix-assisted laser desorption/ionization mass spectrometry (MALDI-MS) imaging. The objectives of the research were to compare HPG-DTX formulations with Taxotere®, an existing commercial treatment, when applied to ex vivo pig bladder samples, which would then be sectioned and examined using MALDI-MS imaging. Results in terms of drug quantitation and penetration in the tissue could then be compared with results obtained using alternative analytical methods.

Methods

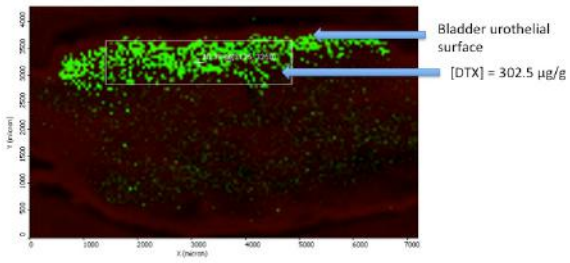
Freshly obtained pig bladders were transferred to pH 7.4 buffer and cut into 1 x 1 cm samples. These samples, urothelium side upwards, were placed in Franz diffusion cells and the donor compartments filled with 300 µls of either an HPG-DTX formulation or a Taxotere® formulation. Following incubation at 37oC, the samples were removed from the cells and frozen before cutting into thin sections at 90o to the urothelium surface. The surfaces of these sections were then examined using MALDI-MS imaging and compared with treated and untreated controls.

Results

The distribution and concentrations of DTX in pig bladder tissue determined using MALDI-MS imaging, as illustrated in the Figure, were comparable with results obtained previously using an established radiolabeled drug method and confirmed the higher loading of DTX in bladder tissue when using an HPG formulation.

Conclusions

The MALDI-MS imaging method provides a convenient means of visualizing DTX in treated bladder tissue and is a potentially useful alternative to the use of radiolabeled drug or other methods for drug quantitation.



Visualization and quantitation of docetaxel in an hpg-dtx-treated pig bladder section using maldi-ms imaging.jpg

Albumin as a Nitric Oxide–Traffic Protein : Novel Anticancer Agent

Friday, 30th September - 16:50 - Targeted drug delivery and Nanocarriers - Nanomedecine for cancer diagnosis & therapy - Amphitheatre 25 - Oral presentation - Abstract ID: 180

Prof. MASAKI OTAGIRI [1], ***Dr. Yu Ishima*** [2], ***Prof. Toru Maruyama*** [3]

1. *Faculty of Pharm Sci., Sojo University,* ***2.*** *Faculty of Pharm Sci , Kumamoto University,* ***3.*** *Faculty of Pharm Sci, Kumamoto University*

Objective While Nitro Oxide (NO) has a wide range of biological functions, NO therapy still has problems that need to be overcome, such as its short half-life in vivo (0.1 s). To accomplish this, we examined human serum albumin (HSA) because it is the most abundant plasma protein and endogenous S-nitrosothiols in human plasma are largely associated with HSA. S-nitrosated HSA (SNO-HSA) is significantly more stable than low molecular weight S-nitrosothiols. It should also be noted that the cellular uptake of NO from HSA with one NO-moiety results in cytoprotective effects, whereas HSA with more SNO-groups induces apoptosis, an effect that is probably useful in cancer therapy. We introduced two type SNO-HSA, ca 7 conjugated SNO groups using chemical modification (Poly-SNO-HSA) and an S-nitrosated recombinant HSA dimer (SNO-AL –Dimer), as possible cancer therapeutic applications.

Results and Discussion The transfer of NO from Poly-SNO-HSA to cells was faster and more pronounced. Surprisingly, the inflow of NO results in apoptotic cell death by ROS induction and caspase-3 activation and not cytoprotection.

Interestingly, NO donors the such as nitroglycerin have been reported to reverse resistant to anticancer agents. Therefore, we evaluated the effect of Poly-SNO-HSA on the resistance of human myelogunous leukemic cells (K562 cells) to doxorubicin (dx). The results showed that treatment with Poly-SNO-HSA increased its accumulation in dx-resistant K562 cells (K562/dx). Furthermore, Poly-SNO-HSA enhanced the anticancer effect of dx in K562/dx mice. Poly-SNO-HSA reversed dx resistance by decreasing the expression of P-gp and HIF-1-α.

The SNO-AL-Dimer was found to specifically deliver large amounts of cytotoxic NO to tumor tissue but not to normal organs in C26 tumor-bearing mice. In this respect, the SNO-AL-Dimer is superior to SNO-HSA and GS-NO. Interestingly, S-nitrosation improved the uptake of the HSA dimer in tumor tissue through augmenting the enhanced permeability and retention (EPR) effect. The data suggest that the SNO-AL-Dimer behaves not only as an anticancer therapeutic drug but also as a potentiator of the EPR effect. Therefore, the SNO-AL-Dimer might be a very appealing carrier for utilizating the EPR effect in cancer therapeutics.

Nanoparticles in proton and heavy ion therapy

Friday, 30th September - 17:15 - Targeted drug delivery and Nanocarriers - Nanomedecine for cancer diagnosis & therapy - Amphitheatre 25 - Oral presentation - Abstract ID: 559

Dr. erika porcel [1], ***Ms. Marta Bolsa*** [1], ***Ms. Daniela Salado*** [1], ***Dr. Lenka Stefancikova*** [1], ***Mr. Vladimir Ivosev*** [1], ***Prof. Sandrine Lacombe*** [1]

1. *ISMO Université Paris Sud/CNRS*

Introduction :Radiotherapy, one of the main treatments in cancer, can be improved by the use of heavy atoms, as radiation enhancers. Many investigations are conducted in this area. The challenge is to increase the radiation damage on tumor whilst preserving healthy tissue by improving targeting. Recent developments in nanotechnology brought new perspectives by using nanoparticles (NP), which can be specifically functionalized.
Methods :We studied, using plasmid probes, the complex molecular damage radio-enhancement induced by NP. The effect at cellular scale was investigated by clonogenic assay. The internalisation and localisation of NP in cells was performed by complementary methods of microscopy (confocal, electronic…).
Results :We have shown, using plasmid probes, that platinum nanoparticles (PtNP) strongly enhance complex molecular damage induced as well by carbon ions [1], as by protons [2] or gamma rays [3]. This effect is not due to the nature of the incoming radiation but explained by the auto-amplification of electron cascades into the nanoparticles. Similar results were found with gadolinium based nanoparticles (GBN) which give the possibility to associate RMN imaging to radiotherapy. Furthermore, a decrease of mammalian cell survival was also observed when GBN or PtNP are associated to ions radiation [4].
Discussion :These results allow us to measure how the use of heavy nanoparticles could improve treatments by enhancing efficiency and targeting of radiations into the tumor. Further assays are in progress to understand the biological mechanisms underlying these effects [5]. New nanoparticles are considered to find the best candidate.
[1] Porcel et al. Platinum nanoparticles: a promising material for future cancer therapy? Nanotechnology 21, 85103 (2010).
[2] Schlathölter et al. Improving proton therapy by metal-containing nanoparticles: nanoscale insights. International Journal of Nanomedicine 11, 1549-1556 (2016).
[3] Porcel et al. Nano-Sensitization under gamma rays and fast ion radiation. J. Phys.: Conf. Ser. 373 (2012).
[4] Porcel et al. The use of theranostic gadolinium-based nanoprobes to improve radiotherapy performances. Nanomedicine 10 (2014).
[5] Stefancikova et al. Effect of gadolinium□based nanoparticles on nuclear DNA damage and repair in glioblastoma tumor cells. Journal of Nanobiotechnology 14 :63 (2016).

Frontier biomedical research: from multi-functional bio-interfaces to biomaterials and tissue engineering

Friday, 30th September - 16:00 - Tissue engineering and regenerative nanomedicine - Tower 24 - Room 101 - Oral presentation - Abstract ID: 503

Prof. Maria Tomoaia-Cotisel [1]

1. *Babes-Bolyai University of Cluj-Napoca, Faculty of Chemistry and Chemical Engineering, 11 Arany Janos Str., 400028 Cluj-Napoca, Romania*

The progress over a quarter century on understanding molecular self-assemblies of various biomolecules, like, fatty acids, lipids, proteins and drugs, and colloidal characteristics of inorganic nanoparticles (NPs), such as hydroxyapatite (HapNPs) as well as of AuNPs and AgNPs, has allowed us to develop a practical strategy for syntheses of innovative nanobiomaterials. These nanobiomaterials can be used as scaffolds in cell culture or in biomedical devices with improved anti-microbial properties. Thus, new horizons are opened up from multi-functional biointerfaces and innovative materials to tissue engineering and nanomedicine with vast biomedical applications. The structure and properties of these materials are investigated by cutting-edge experimental tools existing in our Center of Physical Chemistry, in Babes-Bolyai University platform of research (e.g., AFM, STM, fluorescence microscopy, SEM and TEM, Langmuir-Blodgett techniques for self-assembly (LBT), DSC calorimetry, and various spectroscopic techniques: UV-Vis, FTIR, RMN, RAMAN and RES.

Innovative Hap, modified with Si, Mg and Zn: Hap-Si-Mg-Zn, and functional scaffolds of these materials with collagen:COL represent the first report on the effects of Si, Mg and Zn, simultaneously present within the layered ceramic scaffolds in human osteoblasts culture. For this goal, the cellular expression of osteoblasts markers: like collagen, osteopontin and osteocalcin were visualized by fluorescence microscopy and by using immuno-cytochemical staining methods. Results indicate that combined scaffolds made of Hap/COL, Hap-Si/COL and Hap-Si-Mg-Zn/COL layers have an improved stimulating activity to osteoblasts compared with native scaffolds (e.g., made only from pure Hap), particularly in promoting the formation of mineralized bone matrix. Moreover Hap-Si-Mg-Zn/COL combined layered scaffolds substantially enhanced osteoblasts activity and adhesion, as evidenced by cell expression of collagen, osteopontin and osteocalcin as well as of F-actin stress fibers, in vitro. Thus, current study clearly demonstrated that the incorporation of Si, Mg and Zn within Hap could be an active, safe and inexpensive tool for potential clinical applications in orthopedic surgery, bone cancer therapy and nanomedicine. Acknowledgements: This research was supported by the UEFISCDI through the 171, 241 and 257 projects.

The Sustainable Release of Vancomycin and Its Degradation Products from Collagen/Hydroxyapatite Nano/Micro Structured Layers Prepared using Different Techniques

Friday, 30th September - 16:25 - Tissue engineering and regenerative nanomedicine - Tower 24 - Room 101 - Oral presentation - Abstract ID: 83

Dr. Tomas Suchy [1], ***Dr. Monika Supova*** [1], ***Dr. Eva Klapkova*** [2], ***Dr. Vaclava Adamkova*** [3], ***Dr. Jan Zavora*** [3], ***Dr. Margit Zaloudkova*** [4], ***Dr. Martin Braun*** [1], ***Dr. Rastislav Ballay*** [5], ***Dr. Frantisek Denk*** [1], ***Dr. Jiri Rebicek*** [6], ***Dr. Marek Pokorny*** [6], ***Dr. Pavla Sauerova*** [7], ***Dr. Marie Hubalek Kalbacova*** [7], ***Dr. Lukas Horny*** [8], ***Dr. Jan Vesely*** [8], ***Mr. Tereza Vonavkova*** [8]

1. *Department of Composites and Carbon Materials, Institute of Rock Structure and Mechanics, Academy of Sciences of the Czech Republic, Prague,* ***2.*** *Department of Medical Chemistry and Clinical Biochemistry, Charles University, 2nd Medical School and University Hospital Motol, Prague,* ***3.*** *Clinical microbiology and ATB centre, Institute of Medical Biochemistry and Laboratory Diagnostics, 1st Faculty of Medicine, Charles University in Prague and Motol University Hospital, Prague,* ***4.*** *Department of Composites and Carbon Materials, Institute of Rock Structure and Mechanics, Academy of Sciences of the Czech Republic, Prague, Czech Republic,* ***5.*** *1st Department of Orthopaedics, 1st Faculty of Medicine, Charles University in Prague and Motol University Hospital, Prague,* ***6.*** *Contipro Biotech s.r.o., R&D Department, Dolni Dobrouc,* ***7.*** *Institute of Inherited Metabolic Disorders, 1st Faculty of Medicine, Charles University in Prague, Ke Karlovu 2, Prague,* ***8.*** *Faculty of Mechanical Engineering, Czech Technical University in Prague, Prague*

Introduction

Infections of the musculoskeletal system present a serious challenge in the field of orthopaedic and trauma medicine. The aim of the experiment was to develop a resorbable layer via the controlled elution of antibiotics to be used as a bone/implant bioactive interface particularly in the case of prosthetic joint infections or as a preventative procedure regarding primary joint replacement in a potentially infected site.

Methods

Nano- or micro-structured layers based on collagen (type I, VUP Medical, Czech Republic) and 0, 5 or 15wt% of hydroxyapatite nanoparticles (avg. 150nm, Sigma Aldrich, Germany) were prepared employing the lyophilisation or electrospinning of dispersions with or without 10wt% vancomycin hydrochloride (Mylan S.A.S, France) and subsequently cross-linked by EDC/NHS (Sigma Aldrich, Germany). Pure cross-linked collagen/hydroxyapatite electrospun mats were subsequently impregnated with 10wt% vancomycin. The in vitro release rates of vancomycin and its inactive degradation products were characterized by HPLC. The antimicrobial effects of the layers were determined using agar diffusion testing against four different clinical isolates. The in vitro biological evaluation was conducted using SAOS-2 cells in direct contact with the layers or 24h infusions (MTS/LDH tests).

Results

The maximum concentration of the released active form of vancomycin (700mg/l after 3hours, 150mg/l 21stday) was assessed by means of the vancomycin impregnation of cross-linked electrospun layers. The lowest concen-

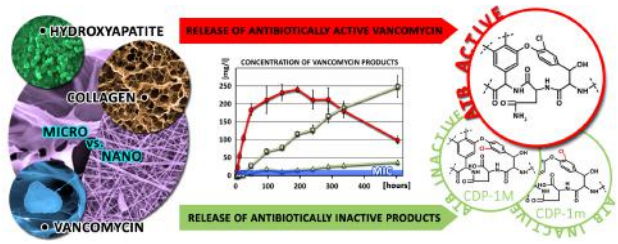
tration was determined for those layers electrospun directly from a collagen solution with vancomycin. Agar diffusion tests revealed that the electrospun impregnated layers exhibited the highest activity. Modification using hydroxyapatite exerts no strong effect on vancomycin evolution. All the tested samples showed a sufficient cytocompatibility rate with no indication of cytotoxic effects.

Discussion

The higher specific surface of nanostructured layers probably plays a negative role in the preparation process due to the higher rate of vancomycin elution to the cross-linking solution. This may be overcome via the subsequent impregnation of the cross-linked layers. Our results suggest that the local application of high-dose vancomycin via drug delivery carriers provides a safe therapeutic osteomyelitis treatment method that prevents the development of bacterial resistance.

Acknowledgments

This study was supported by the Technology Agency of the Czech Republic (project no. TA04010330).



Iconan suchy.jpg

Gold Nanoshell-Assisted Wireless Activation of Myotube Contraction

Friday, 30th September - 16:50 - Tissue engineering and regenerative nanomedicine - Tower 24 - Room 101 - Oral presentation - Abstract ID: 547

Mr. Attilio Marino [1], ***Dr. Satoshi Arai*** [2], ***Dr. Yanyan Hou*** [2], ***Prof. Madoka Suzuki*** [2], ***Prof. Gianni Ciofani*** [1]

1. *Istituto Italiano di Tecnologia,* ***2.*** *Waseda University*

INTRODUCTION

Different approaches have been developed in the recent years for muscle cell stimulation and for myotube contraction, aiming at various applications in tissue engineering, regenerative medicine, and bionics. Mild heat stimulation of muscle cells within physiological range represents an intriguing strategy for the modulation of cell functions. In this context, plasmonic properties of gold nanostructures can be exploited upon near-infrared (NIR) excitation in order to remotely heat cells, also into deep tissues, owing to the low absorption in the NIR (λ 800 nm) [1].

Although the effects of nanoparticle-assisted photo-thermal stimulation have been widely investigated in neurons and nerves, no significant studies on muscle cells can be found in the literature. In this work, for the first time, photo-thermal conversion was exploited to remotely stimulate muscle cells by using gold nanoshells (NS) in combination with NIR radiation.

MATERIALS AND METHODS

Au@SiO2 NS in water suspension (50 μg/ml, Nano-Composix) were centrifuged and re-dispersed in the cell medium (50 μg/ml). NS were characterized with scanning electron microscopy and UV-Vis absorbance was also investigated (Hitachi F-2700).

Intracellular temperature dynamics in response to a NIR stimulation (0.25 W/cm2) were assessed both in presence and in absence of the NS (Nano-Composix, 50 μg/ml) treatment, after 5 days of differentiation, by using a temperature-sensitive fluorescent thermometer targeting endoplasmic/sarcoplasmic reticulum (ER thermo yellow).[2] Calcium imaging was performed by using Fluo-4 AM fluorescent indicator.

RESULTS AND DISCUSSIONS

Intracellular temperature increments of about 5°C were induced by the NIR stimulation in the presence of NS. The increments of temperature were demonstrated able to efficiently induce a myotube contraction. Fluorescence Ca2+ imaging analysis demonstrated as no intracellular Ca2+ transients can be detected during the NIR + NS stimulation, so indicating that Ca2+ fluxes are not involved in the described phenomena.

Our results report, for the first time, a "wireless" activation of muscle cells mediated by light and nanoparticles, envisaging applications in tissue engineering as well as in bionics.

REFERENCES

[1] J. Yong et al. Adv. Healthc. Mater. 2014, 3, 1862–186.

[2] S. Arai et al. Sci. Rep. 2014, 4.

Complete 3D regeneration of functional organs by manipulation of the pluripotent stem cells

Friday, 30th September - 17:15 - Tissue engineering and regenerative nanomedicine - Tower 24 - Room 101 - Oral presentation - Abstract ID: 47

Prof. Hiroshi Kagami [1]

1. *Shinshu University*

Complete regeneration of the functional organs have been very difficult in many biological species. We have been challenged the regeneration of the functional organs by use of the pluripotent stem cells isolated from early avian embryos. These stem cells were cultured, genetically modified and micro-injected into the other avian embryos. Completely functional legs and hearts could be regenerated and the organ shape in 3D level were almost same as that of the control organs. Moreover, the mice ES cells could be committed into avian embryos and the mice derived neuron or muscle could be regenerated in the avian embryos.
All of the experimental procedures and results of the regeneration of the stem cell-derived organs will be shown as movie.

Evaluation of theranostic dendrimers for radiotherapy and MRI of gliomas

Friday, 30th September - 16:00 - Nanomedecine for cancer diagnosis & therapy - Tower 24 - Room 103 - Oral presentation - Abstract ID: 466

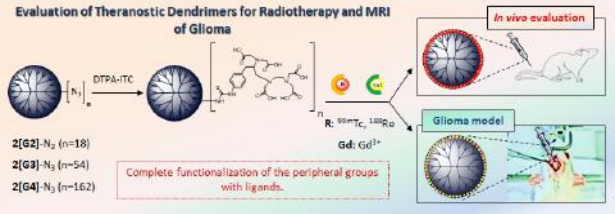
Ms. Flonja Liko [1]***, Prof. Eduardo Fernandez-Megia*** [2]***, Prof. Francois Hindré*** [1]

1. *Micro et Nanomédecines Biomimétiques (MINT) UMR-S 1066, IBS-CHU, University of Angers, Angers,* ***2.*** *Center for Research in Biological Chemistry and Molecular Materials (CiQUS), University of Santiago de Compostela, Santiago de Compostela.*

Aim: A new oncologic strategy, based on the integration of nanovectorized radiotherapy and locoregional delivery was evaluated for the treatment of gliomas. Our focus was the synthesis of heterofunctional dendrimers to facilitate personalized management of the most common and lethal type of primary brain tumors. Gallic acid-triethylene glycol (GATG) dendrimers, fully functionalized with DTPA derivatives, were the nanovectors of choice to deliver the radiotherapeutic 188-Re and paramagnetic nuclei Gd(III), targeting the radiotherapeutic dose to the tumor site with a minimally invasive stereotactic surgery, in an F98 rat glioma model. Intravenous injection was used to further investigate the pharmacokinetics, throughout body distribution and clearance profiles of these dendrimers.

Methods: F98 tumor cells were implanted intracerebrally in syngeneic Fischer rats (n=15). A comparison of brain retention and tissue biodistribution between 188Re-perrhenate (n=6) and (188Re+Gd)-2[G3]-DTPA dendrimers (n=6) was done following CED (3.7 MBq/injection) at Day 20 post-tumor implantation, using an osmotic pump (0.5µL/min for 20 minutes). Brain retention of Gd-2[G3]-DTPA dendrimers in F98 glioma rats, was also evaluated by MRI, at 24h post-CED (n=3). Intravenous injection was used to mimic the behavior of dendrimers in the bloodstream, following their brain clearance. To investigate if the generation sizes of dendrimers influenced any differences in their pharmacokinetics, throughout body distribution and clearance profiles, three highest generations (2[G2]-2[G4]) of GATG dendrimers were synthesised and radiolabeled with technetium. Rats (n=36) were intravenously injected with 3.7 MBq of 99mTc-2[G2]-DTPA, 99mTc-2[G3]-DTPA, and 99mTc-2[G4]-DTPA dendrimers, and put in individual metabolic cages. They were sacrificed at 1, 6, 24 and 48 hours post-injection (n=9 per each time interval; 3 rats per generation of dendrimers). The content activity of urine, feces and each organ was determined using a gamma counter and expressed as percentage of injected dose/organ (%ID).

Conclusion: The use of dendrimers as nanovectors prevented the fast brain clearance of the radionuclide on its own, and prolonged the confinement of the internal radiation at the tumor site. Molecular weight and architecture of dendrimers had an important role on the in vivo behavior of these nanovectors. The easy dual labeling procedure opens the perspective of multimodality in therapy and imaging of gliomas.



Gatg dendrimers for integrated radiotherapy and mr imaging of glioblastoma.jpg

Targeting Hypoxia in Advanced Prostate Cancer Using Tirapazamine-Copper Nanoparticles

Friday, 30th September - 16:25 - Nanomedecine for cancer diagnosis & therapy - Tower 24 - Room 103 - Oral presentation - Abstract ID: 253

Dr. Wafa Al-Jamal [1]

1. *University of East Anglia/ School of Pharmacy*

Introduction: Hypoxia is considered a hallmark of cancer and a common characteristic of locally advanced solid cancers, including prostate cancer. Tumor hypoxia plays a key role in promoting angiogenesis, metastasis, and drug resistance. Hypoxia-activated prodrugs have opened the door for specific treatment of the solid and metastatic tumors with lower side effects. Tirapazamine (TPZ) is the most advanced hypoxia-activated prodrug. TPZ has shown great specificity and potency in inhibiting tumor growth at moderate to severe hypoxic conditions. TPZ is currently in phase III clinical trials to treat cervical cancer, however its efficacy in vivo has been limited due to its poor penetration in tumor tissues. The present work aims to develop novel TPZ-copper nanoparticles to target and penetrate hypoxic prostate tumor mass, following systemic administration in vivo.
Methods: To improve the encapsulation of TPZ in nanoparticles, TPZ-copper complexes, Cu(TPZ)2, were prepared and characterized using different analytical techniques (HPLC, IR, UV/Vis, spectrofluorometry and MALDI-TOF). Next, a remote loading method was developed to encapsulate stable Cu(TPZ)2 complexes in different lipid-based nanoparticles. Cu(TPZ)2–nanoparticles were characterized using dynamic light scattering, spectrofluorometry and HPLC. The cytotoxicity of TPZ and Cu(TPZ)2 complexes, and Cu(TPZ)2–nanoparticles was assessed in vitro, using prostate cancer cells cultured under normoxia and hypoxia. The cytotoxicity was evaluated using resurazin cell viability assay.
Results: Cu(TPZ)2 complexes were more hydrophobic in nature, which led to an efficient drug loading in the lipid nanoparticles. The drug loading was dependent on the lipid composition, drug: lipid and the hydrating buffers used. Our results showed that the cytotoxicity of the Cu(TPZ)2–nanoparticles was highly selective to hypoxia. It was also dependent on the cell line, drug concentration, and the incubation time.
Conclusions: In this work, we showed that the complexation of TPZ with copper was an attractive approach to promote TPZ encapsulation in lipid nanoparticles. This novel nanomedicine could overcome TPZ shortcomings in vitro and in vivo, and offer a promising approach to target advanced prostate cancer in patients.
Acknowledgements: This work was supported by Prostate Cancer UK (Grant CDF-12-002), the Engineering and Physical Sciences Research Council (EPSRC) (EP/M008657/1), and University of East Anglia.

Theoretical Reactivity of Cell-Surface Receptors in Presence of Nanodiamonds as Carriers

Friday, 30th September - 16:50 - Nanomedecine for cancer diagnosis & therapy - Tower 24 - Room 103 - Oral presentation - Abstract ID: 433

Dr. Norma Flores-Holguin [1], ***Prof. Linda-Lucila Landeros-Martinez*** [1], ***Dr. Erasmo Orrantia-Borunda*** [1]

1. *Centro de Investigación en Materiales Avanzados,S.C.*

Introduction

The estrogen receptors (ER) have allowed offering a better prognosis to patients with breast cancer. The most common treatment is Tamoxifen (TAM), a Selective Estrogen Receptor Modulator (SERM), which can inhibit the estrogen effects in the breast cancer neoplastic cells without altering the beneficial effects it has on bone, cardiovascular and nervous system [1]. However, even though TAM has good results, it also has side effects such as blood clots and endometrial cancer.

For this reason a study of the theoretical reactivity parameters of two cell-surface receptors, arginine-glycine-aspartic acid tripeptide (RGD) that is present in the glycoprotein fibronectine and Aspargine-Glycine-Arganine tripeptide (NGR) found in the aminopeptidase, was developed trying to find their interaction with TAM drug.

Methodology

The computational characterization of the peptides RGD and NGR, and the complex ND-TAM/RGD, ND-TAM/NGR, and ND-TAM complex was made using the Conceptual Density Functional Theory, DFT [2] with M06 functional [3] and 6-31G (d) basis set [4]. The reactivity parameters as ionization potential, electron affinity and chemical hardness were calculated. The cross reactivity was done and the frontier orbitals were obtained for all the studied systems.

Results and Discussion

According with electronic density distribution, the ND-TAM complex inactivates the drug on its surface. This does not permit to bind it with the cell-surface receptors in healthy cells. Nevertheless, the difference in pH of cancer cells will allow the binding in the active site of the receptor. The analysis of the frontier molecular orbitals confirm the inactivation of TAM when it is carried by the nanodiamond. It can be observed in Figure 1. Also, the cross-reactivity indicated that TAM reacts with greater facility in presence of RGD than NGR.

Keywords: breast cancer, Tamoxifen, Estrogen Receptor, DFT

[1] F. Cammarata-Scalisi1, M. Balza, A. Arenas de, M.M. Sotolongo, F. Stock, J.L. Valderrama-Landaeta, Revista Médica de la Extensión Portuguesa, 2 (2008)

[2]. P. Hohenberg, W. Kohn, Physical Review, 136 (1964) B864-B871.

[3] Y. Zhao, D. Truhlar, Theoretical Chemistry Accounts, 120 (2008) 215-241.

[4] V.A. Rassolov, M.A. Ratner, J.A. Pople, P.C. Redfern, L.A. Curtiss, Journal of Computational Chemistry, 22 (2001) 976-984.

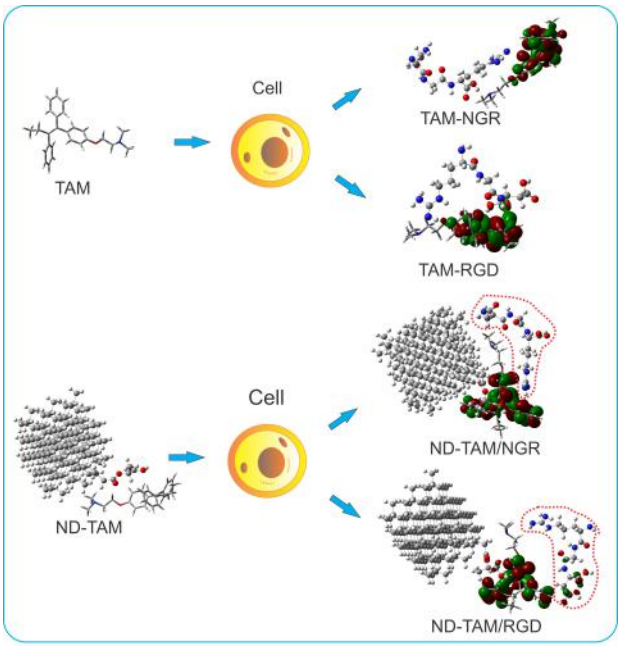


Figure 1.jpg

Photocatalytic TiO2 Nanoparticles for Tumor Therapy

Friday, 30th September - 16:00 - Nanomedecine for cancer diagnosis & therapy - Tower 24 - Room 105 - Oral presentation - Abstract ID: 56

Ms. Susanne Koch [1], ***Dr. Sofia Dembski*** [2], ***Dr. Stephan Hackenberg*** [3], ***Dr. Karine Heuzé*** [4]

1. *University of Bordeaux / Fraunhofer ISC Würzburg,* ***2.*** *Fraunhofer ISC Würzburg,* ***3.*** *Department of Oto-Rhino-Laryngology, Plastic, Aesthetic and Reconstructive Head and Neck Surgery, University Clinic Würzburg,* ***4.*** *University of Bordeaux*

Despite advances in treatments for head and neck cancers, patient survival rates have only slightly increased over the last decades [1]. Thus, novel therapy strategies need to be developed to enhance current standard treatments like surgery, radiation or chemotherapy. Photocatalytic nanoparticles (NPs) are a serious candidate as they may combine their function as a therapeutic agent with surfaces that can be functionalized to mediate cellular uptake, carry drugs or molecules for diagnostic.

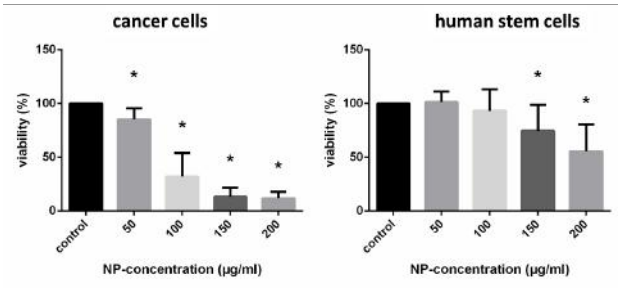
In this study, TiO2 (anatase) NPs with diameters of 10 nm were synthesized via hydrothermal treatment. The NPs and their in situ grafted organic surface groups were characterized by XRD, TEM, zeta potential, dynamic light scattering (DLS), thermogravimetric analysis, photocatalytic activity measurements and IR-spectroscopy. Stabilization of the synthesized NPs in cell culture media was examined via sedimentation studies (absorption spectroscopy, DLS). Cytotoxicity tests and tumor treatment experiments were carried out in a human tumor cell line (FaDu) and human bone marrow-derived mesenchymal stem cells with the help of the MTT colorimetric staining method.

After complete characterization of the synthesized TiO2 NPs, they were stabilized in cell culture media using a polycarboxylate ether as a novel stabilization agent. A perfect stability over at least one day and agglomerate sizes less than 100 nm were achieved [2]. Within this research the biocompatibility of non-activated NPs was demonstrated. For the treatment of tumor cells a novel concept was developed; the photocatalytic NPs were activated by UV light which modified their surface groups. Hereupon, NPs exhibited dose-dependent cytotoxic properties in FaDu while non-malignant cells were significantly less affected.

In summary, the novel NPs are attractive for the treatment of superficial mucosa malignancies of the head and neck due to their cancer specific toxicity and since they can be activated prior to cell exposure. This allows tumor therapy without UV irradiation of cells avoiding UV-induced DNA damages in non-targeted cells. Furthermore, pre-treatment UV activation allows the NP application in deeper cancer formations.

[1] R. Siegel et al. CA Cancer J. Clin. 65 (2015) 5–29.

[2] S. Koch et al. Colloids Surf. B: Biointerfaces 143 (2016) 7-14.



Vitality of cancer cells and human stem cells treated with pre-activated tio2 nanoparticles.png

Immobilized ERK2 and GSK-3beta magnetic microparticles for targeted protein phosphorylation

Friday, 30th September - 16:25 - Nanomedecine for cancer diagnosis & therapy - Tower 24 - Room 105 - Oral presentation - Abstract ID: 299

Dr. Marcela Slovakova [1], ***Ms. Lenka Hromádková*** [2], ***Mr. Rudolf Kupčík*** [1], ***Dr. Daniela Řípová*** [3], ***Prof. Zuzana Bílková*** [1]

1. *University of Pardubice, Faculty of chemical technology,* ***2.*** *National institute of mental health, Department of Neurobiology and AD center, Klecany. Charles University in Prague, Faculty of Science, Prague,* ***3.*** *National institute of mental health, Department of Neurobiology and AD center, Klecany*

Introduction: Micro and nanoparticles with immobilized enzymes as recoverable, stable and specific catalysts are applied in a variety of technologies, in biomedical applications and widely in research. Magnetic microparticles bring the advantage of the non-contaminating and very specific and sensitive reaction on their substrates, peptides and proteins. This work is based on the use of proline-directed protein kinases: extracellular signal-regulated kinase (ERK2) and glycogen synthase kinase 3β (GSK-3β) immobilized to magnetic microparticles for targeted recombinant tau 1-441 phosphorylation.

Methods: In order to obtain highly efficient carrier for the targeted phosphorylation of peptides/proteins, two kinases ERK2 and GSK-3β were immobilized to various superparamagnetic beads with carboxylic, aldehyde, and metal cations Ni^{2+} or Co^{3+} functionalities. Relevant methods of covalent immobilization, non-oriented and oriented, were chosen and reaction conditions were optimized. Phosphorylation of low molecular peptides and operational and storage stabilities of kinase-superparamagnetic particles were performed. Soluble and immobilized ERK2 and GSK-3β were applied for recombinant tau 1-441 phosphorylation. Tryptic phosphopeptides enrichment was performed by ion-metal affinity chromatography using TiO_2 magnetic nanoparticles. The level and the position of phosphorylation sites were identified by using MALDI-LTQ-Orbitrap MS. Western blot was carried out to confirm phosphorylation of tau 1–441 (ENZO and rPeptide) by soluble or immobilized ERK2 and GSK-3β kinases using specific anti-tau and anti-phospho-tau antibodies.

Results: The effect of immobilization was confirmed due to the enzymes ability to phosphorylate low molecular synthetic peptides. The ERK2 and GSK-3β kinases immobilized on SeraMag beads using the carbodiimide chemistry proved the ability to phosphorylate peptides in 10 cycles without significant loss in activity. Using carriers with immobilized ERK2 or GSK-3β we performed the efficient phosphorylation of model recombinant protein tau 1-441. Presence and position of phosphorylation along the peptide chains were detected by MALDI MS and confirmed also by Western blot with anti-tau phospho-specific antibodies.

Discussion: Present results document very well the ability of ERK2 and GSK-3β superparamagnetic particles to phosphorylate the target peptides and recombinant tau 1-441 with desired efficiency and specificity. The process of phosphorylation can be better controlled; subsequent purification of phosphorylated tau can be omitted.

Acknowledgement: EU project NADINE No. 246513

Evaluation of magnetic nanoparticles coated by Taxol imprinted polymer for controlled drug delivery in mouse breast cancer model

Friday, 30th September - 16:50 - Nanomedecine for cancer diagnosis & therapy - Tower 24 - Room 105 - Oral presentation - Abstract ID: 468

Prof. Hamid Hashemi-Moghaddam [1], ***Mrs. Elnaz Mirsaeed-Gazi*** [1], ***Prof. Saeed Zavareh*** [2]

1. *Damghan Branch, Islamic Azad University,* ***2.*** *Damghan University*

Nanoparticles (NPs) have been extensively investigated to improve delivery efficiency of therapeutic and diagnostic agents. In this study, magnetic molecularly imprinted polymer (MIP) was synthesized by using poly DOPA. Synthesized MIP was used for controlled Taxol delivery in a spontaneous model of breast adenocarcinoma in Balb/c mice in the presence of an external magnetic field. Antitumor effectiveness of Taxol imprinted polymer (Taxol-IP) was evaluated in terms of tumor-growth delay, tumor-doubling time, inhibition ratio, and histopathology. Results showed higher efficacy of Taxol-IP in the presence of magnetic field upon suppressing tumor growth than free Taxol and Taxol-IP without magnetic field. The Taxol and Fe distribution among tissues were evaluated by high-performance liquid chromatography and flame atomic absorption spectrometry, respectively. The obtained results, showed significantly deposition of Taxol in the Taxol-IP treated group with magnetic field. Thus, magnetic Taxol-IP is promising for breast cancer therapy with high efficacy.

Enhanced antifungal activity of itraconazole by the supercritical antisolvent technique

Friday, 30th September - 16:00 - Targeted drug delivery and Nanocarriers - Tower 24 - Room 107 - Oral presentation - Abstract ID: 304

Dr. Jayvadan Patel [1]

1. *Faculty of Pharmacy, Sankalchand Patel University, Visnagar, Gujarat, India*

Itraconazole is broad spectrum antifungal drug that have problems of poor solubility and bioavailability of conventional dosage forms. Attempts to overcome the solubility problems are solubilisation with mixed micelles or forming a complex using cyclodextrins but these approaches are of limited success. One tried to improve the in vivo performances of poorly soluble drug by reducing the particles size using supercritical antisolvent technique (SAA). Micronization of itraconazole dissolved in acetone, dimethyl sulfoxide and ethanol with supercritical carbon dioxide as antisolvent was successfully performed using a supercritical antisolvent technique. The effect of a few process parameters such as precipitation temperature, the pressure and solute concentration in the liquid solution has been studied to evaluate their influence on morphology and size of particles. The micronised itraconazole were evaluated for drug content, particle size analysis and in vitro dissolution profiles. Fourier transform infrared spectroscopy, differential scanning calorimetry and PXRD patterns was used to study the possible changes after micronization of itraconazole. The dissolution rate was increased after micronized compared with pure itraconazole in distilled water, pH 1.2 buffer and pH 7.0 buffer. Micronized and pure itraconazole were evaluated for their therapeutic efficacy in the treatment of experimental oral candidiasis induced by Candida albicans in immunosuppressed rats. This antifungal activity was analyzed by microbiological and histopathological techniques. Microbiologically, micronized itraconazole significantly ($p<0.05$) reduced the number of colony forming units (CFU) sampled from the oral cavity of rats treated for eight consecutive days, compared to untreated control rats. Histologically, the untreated control animals showed numerous hyphae on the epithelium of the dorsal surface of the tongue. In contrast no hyphal colonization of the epithelium was seen in micronized itraconazole-treated animals indicating improved bioavailability of micronised itraconazole as compared with pure itraconazole. In conclusion, SAA process could be a useful method for the micronization of itraconazole and its solubility, dissolution rate and antifungal activities were significantly increased by micronization. Improve the in vivo performances of poorly soluble drug by reducing the particles size using supercritical antisolvent technique (SAA) is a novel approach.

WORKSHOP: Synthesis of amphiphilic hyaluronan containing-phenyl fatty acids for the preparation of polymeric micelles for applications in drug delivery

Friday, 30th September - 16:25 - Targeted drug delivery and Nanocarriers - Tower 24 - Room 107 - Workshop presentation - Abstract ID: 68

Dr. Gloria Huerta-Angeles [1], ***Ms. Alena Matelova*** [2], ***Mrs. Zdislava Brunová*** [1], ***Dr. Daniela Šmejkalová*** [1], ***Prof. Vladímir Velebný*** [1]

1. *CONTIPRO,* ***2.*** *Department of Chemistry, Faculty of Technology, Tomas Bata University in Zlín*

Introduction

Polymeric micelles (PM) are particularly interesting for drug delivery applications because of their characteristic features e.g., size, surface, solubility under physiological conditions and enhanced permeability, which help them to penetrate and be taken up by cells. Hyaluronan (HA) is a natural polysaccharide, ubiquitous in the human body, therefore, it is suitable for medical applications. Moreover, HA allows specific targeting.

Methods

In this work, novel amphiphilic hyaluronan (HA) based polymeric micelles were prepared in base of HA grafted with omega-phenylalkanoic acids (ω-PAA), including 4-phenylbutyric, 6-phenylhexanoic, 8-phenyloctanoic or 11-tolylundecanoic acids, which are aromatic fatty acids normally occurring in fats of natural origin. The prepared derivatives were encapsulated of aromatic hydrophobic drugs in order to evaluate its loading capacity (ie. trans-resveratrol, retinyl palmitate and quercetine).

Results

The synthesis of HA-grafted with ω-PAA (HA-g-PA) was mediated by mixed anhydrides1 that allows HA modification without degradation or formation of toxic subproducts. The reactivity of ω-PAA towards esterification has decreased with the increasing length of the aliphatic spacer between the aromatic ring and carboxylic moiety. The novel HA derivatives were found to be not cytotoxic and self-assemble from very low CMC. Furthermore, Polymeric micelles (PM) were characterized by small size (cca 30 nm, PDI; 0.2).

Discussion

Comparing to our previously reported work2, the drug loading capacity of HA-g-PA. The drug loading content increased substantially likely because of pi-pi interactions between the micelle core and loaded aromatic hydrophobic drug. The formation of well-defined hydrophobic nanodomains can be reached by defining the degree of modification of HA. This work describes a facile strategy to achieve well defined HA derivatives possessing an effective drug loading content, which is a critical factor for designing polymeric micelles3.

References

1. Huerta-Angeles, G., et al., Carbohydrate Polymers, 2014, 111, 883-891.
2. Šmejkalová, D., et al., Biomacromolecules, 2014, 15(11), 4012-4020.
3. Liang, Y et al., Biomaterials, 2015, 71, 1-10.

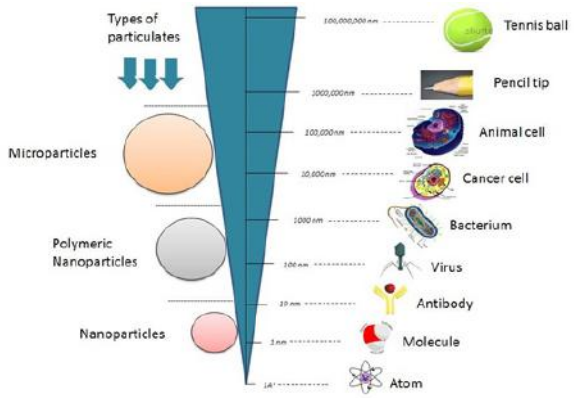
Effects of nanoparticles on gastrointestinal disorders and therapy

Friday, 30th September - 18:15 - Video Presentations - premc.org - Video presentation - Abstract ID: 217

Ms. Rabia Riasat [1], ***Prof. Nie Guangjun*** [1], ***Ms. Zertashia Tariq*** [2]

1. *CAS Key Laboratory for Biomedical Effects of Nanomaterials and Nanosafety, National Center for Nanoscience and Technology, Beijing, 100190, China.,* ***2.*** *Department of Biological Sciences, Islamia University, Bahawalpur, Pakistan.*

Gastrointestinal (GI) diseases are the diseases that affect any part of the gastrointestinal tract including acute, chronic, recurrent or functional disorders. There are a numbers of factors affecting the biology of GI tract. Nanoparticles are one of them in causing diseases. As there are many conventional therapeutic strategies for the treatment of GI diseases but these are not very efficient. Nanotechnology is the emerging and rapidly evolving field of the current era with new hopes in the field of nano-medicine for the detection, prevention and the treatment of diseases. Chemotherapeutic drug delivery in the field of nanotechnology has gained much attention and focus recently. Nano materials have wider range of potential applications for the detection and treatment of diseases while toxicological effects cannot be neglected and safe and non-toxic nano drugs should be considered for the treatment of pathological and physiological gastrointestinal diseases to reduce the existing conventional treatments. The parameters such as shape, size, surface chemistry and geometry of nanoparticles are important to consider in the designing of nano carrier. The review aims at integrating toxicological effects of nano materials and their safe and effective role in the treatment of GI disorders. Although there are disorders caused by nanoparticles but counteracting as well in a safe and targeted delivery of the conventional drugs into the GI system.



Img1.jpg

Synthesis and characterization of biocompatible, non-toxic dopamine coated novel flower shapes core (Fe)/ porous hollow shell (Fe3O4) super paramagnetic Fe/Fe3O4 nanoparticles

Friday, 30th September - 18:30 - Video Presentations - premc.org - Video presentation - Abstract ID: 263

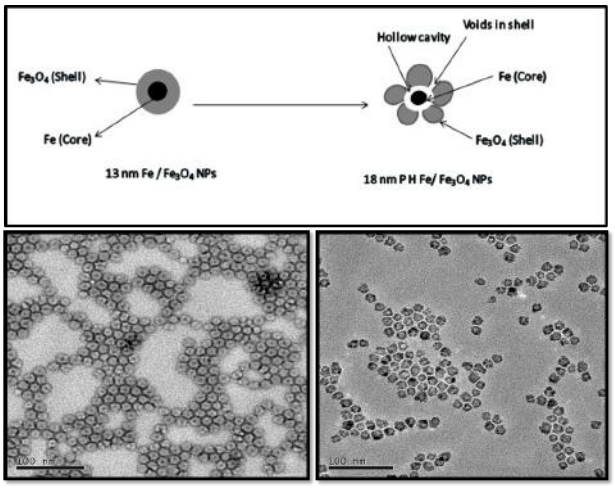
Ms. Rabia Riasat [1]***, Prof. Nie Guangjun*** [1]***, Ms. Zertashia Tariq*** [2]***, Ms. Muntaha Sakeena*** [3]

1. *CAS Key Laboratory for Biomedical Effects of Nanomaterials and Nanosafety, National Center for Nanoscience and Technology, Beijing, 100190, China.,* ***2.*** *Department of Biological Sciences, Islamia University, Bahawalpur, Pakistan.,* ***3.*** *EME, NUST, Islamabad, Pakistan*

Superparamagnetic flower shaped Fe/Fe3O4 nanoparticles with iron core and hollow and porous shell were synthesized by thermal decomposition of iron penta carbonyl. Controlled oxidation of these nanoparticles was done by acid etching. Fe/Fe3O4 core shell iron oxide nanoparticles were synthesizing first as the template material at annealing temperature of 120˚C and solution based conversion of Fe/PHFe3O4 nanoparticles was done through two way annealing progression of heating time, first at 100˚C then at 250˚C with constant heating rate of 5˚C/min. Phase confirmation of these nanoparticles were done by X-ray diffraction (XRD) and morphological structures were analysed by transmission electron microscopy (TEM) and higher resolution transmission electron microscopy (HRTEM) and showed that nanostructures were approximately in 16 nm to 20 nm in size. While the opening pores in the shell were 1-3nm in size and the hollow cavity between iron core and shell is about 2nm hollow and porous Fe/Fe3O4 nanoparticles were surface functionalized by dopamine polymer. The dopamine coating was confirmed by Fourier transform infrared (FTIR) spectra. Nanoparticles showed magnetic saturation of 38 emu/g analysed by vibrating sample magnetometer (VSM) and showing the magnetic property as well after coating with dopamine. The as prepared nanoparticles are hydrophilic to aqueous media and are not toxic to cells (Caco-2 and Hep G2).These nanoparticles can be exploited for a number of applications including target definite therapeutic applications by encapsulating the required drug according to disease profile and can be used a nanomedicine for the iron deficiency control as by possessing large surface area and nano-sized particle profile.

Key words

Core shell nanoparticles, SPIONS, super paramagnetic iron oxide nanoparticles, dopamine



Rabia paper figure.png

Competitive intelligence study of a non-viral vector for gene therapy of Parkinson´s disease (Cinvestav-PX-001)

Friday, 30th September - 18:45 - Video Presentations - premc.org - Video presentation - Abstract ID: 439

Dr. AMERICA PADILLA [1], ***Dr. Daniel Martínez-Fong*** [1], ***Dr. Victor Manuel Téllez-López*** [1], ***Dr. Armando De Jesús Espadas-Álvarez*** [1]

1. *Centro de Investigación y de Estudios Avanzados del IPN*

Introduction

Cinvestav-PX-001 is a nanoparticle-based gene delivery system capable of targeting therapeutic genes to dopaminergic neurons in the brain and therefore induces a site-directed therapy for Parkinson's disease (PD). Experiments in rats with PD-like lesions suggest that this treatment helps to restore dopaminergic function and improve motor symptoms. Cinvestav-PX-001 may have significant commercial potential. This new treatment is safer compared to more commonly used viral vectors for therapeutic indication of PD, because they can induce adverse side effects. Competitive advantages include: high neuronal specificity in addressing the long-term transgene expression, safety and a simple, reproducible and cost-effective synthesis. The aim of this study was to obtain the competitive outlook of the available technologies related with the use of a non-viral vector for gene therapy of PD and thus provide support for strategic decisions in our organization based on the position of intellectual property.

Methods

A prior art analysis based on patents and scientific documents was performed to identify potential competing patents. We applied analysis tools for designing commercialization strategy and these include: a) technological and competitive intelligence, b) market research (size, segments, trends, drivers and growth potential) and c) regulatory environment, in which the technology competes. These aspects were analyzed in order to improve the timeliness and quality of inputs to the decision process.

Results and Discussion

The invention is directed to the market of supplies for gene therapy, the projected growth for the market is up to 2.8 trillion on 2024, and the main leading companies are located in United States, Japan, United Kingdom, France, Germany, Italy and Spain. As a result of this analysis, the commercialization strategy for Cinvestav-PX-001 considers provide a non-exclusive license to an international licensee in USA and/or Europe with the experience and ability to complete its commercial development (clinical phase and regulatory affairs) and the creation of a spinoff company from Cinvestav with innovative vision in new therapeutic applications of Cinvestav-PX-001.

Project supported by Fund of Innovation (Finnova 224222), Ministry of Economy and Conacyt Mexico. We appreciate the support of ISIS Enterprise and Newton Fund in IP analysis.

Diapositiva1.jpg

Authors Index


PremC
Conferences, Events & Workshops