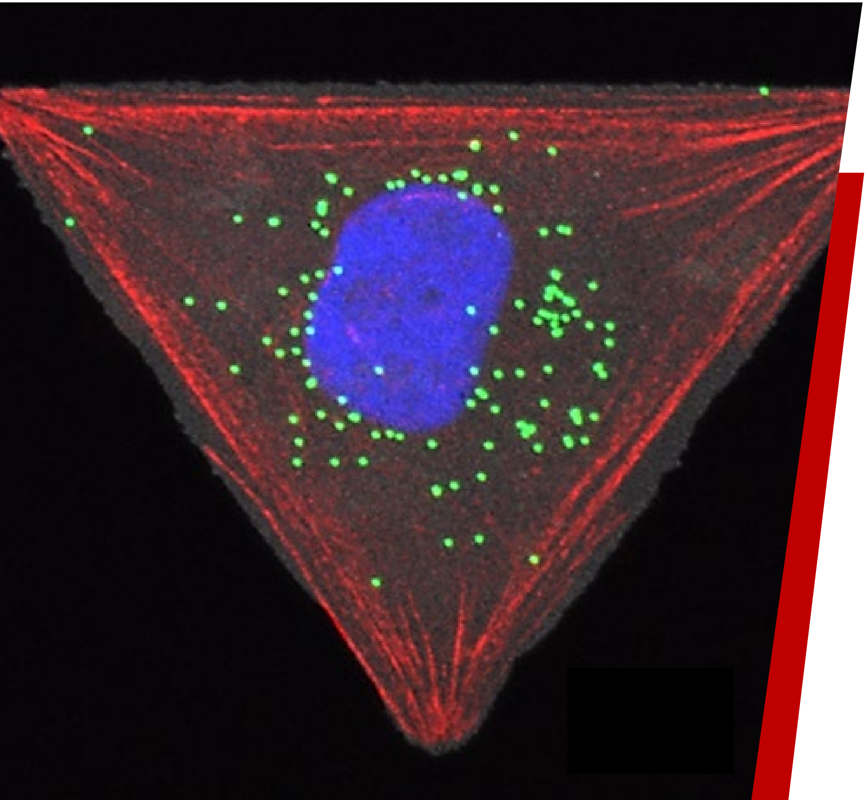


Nanomaterials-cell interactions

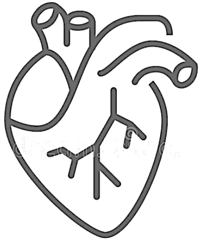
Prof. Dr. Peter Wick

Head Particles-Biology Interactions, Empa

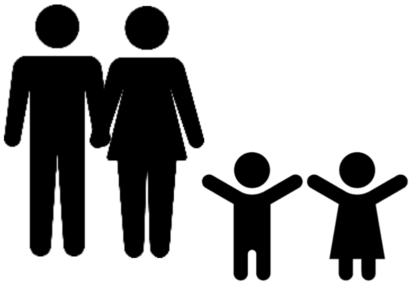
Summerschool, August 19-23 2024, Visp, CH

Nanomaterial safety research along increasing biological complexity

cell



organ



human population

One Health

acute/single dose

freshly synthesized

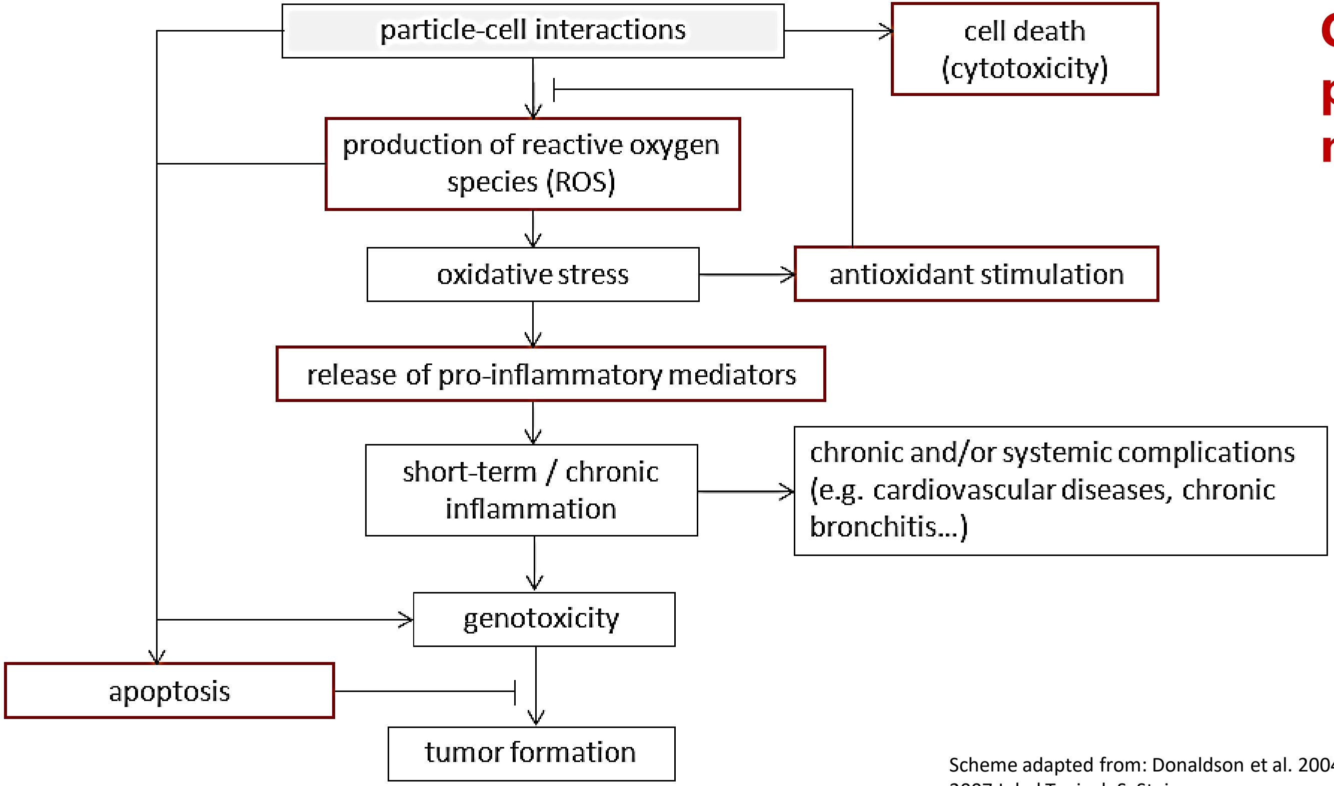
repeated dose

processed

long-term

end-of-life

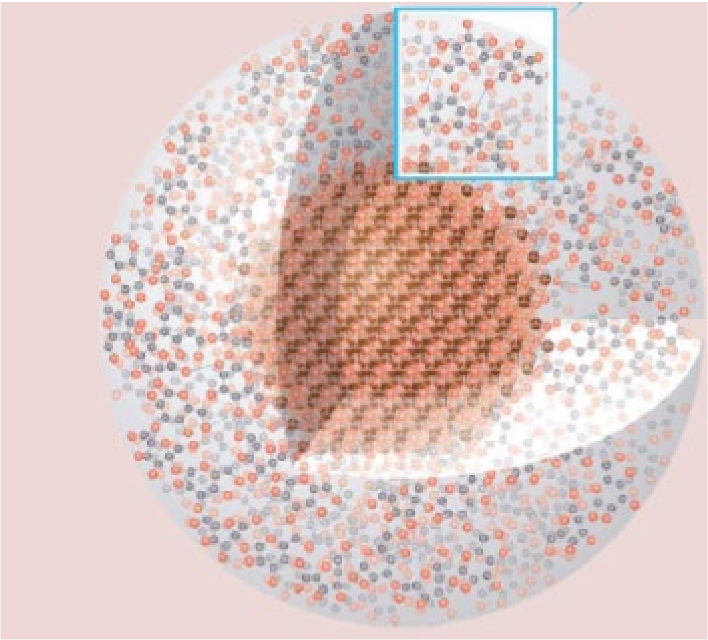
We can build on three decades of anthropogenic nanomaterial tox-research

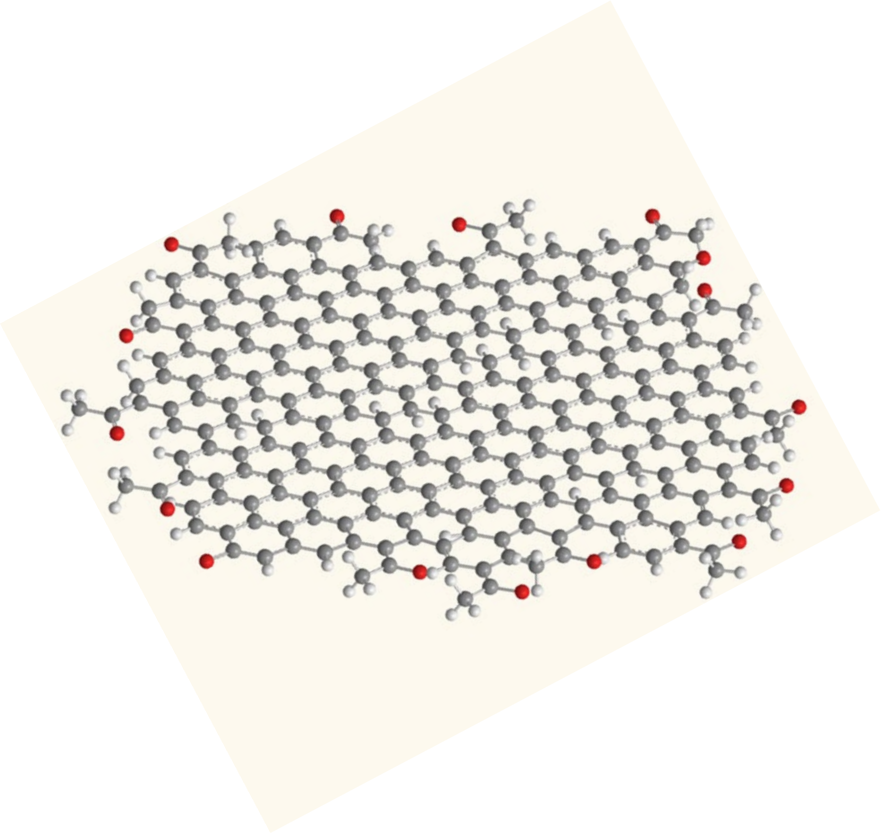


Oxidative stress paradigm for nanomaterials

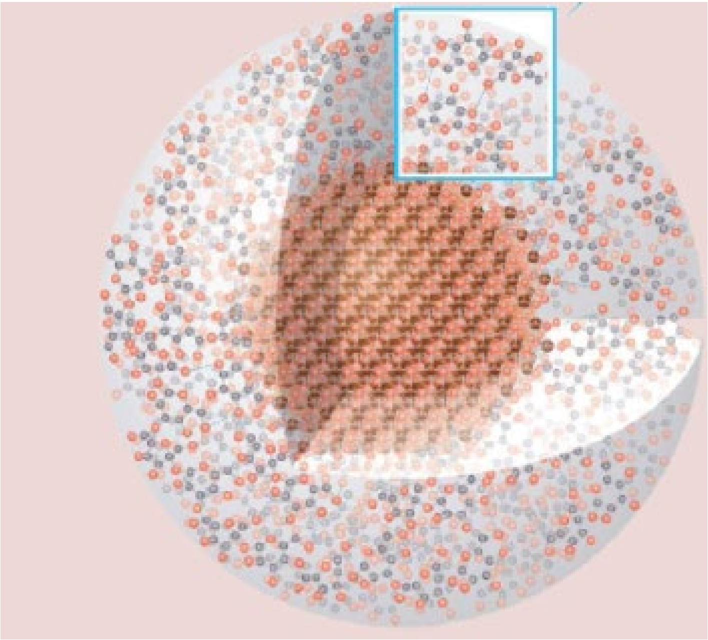
Scheme adapted from: Donaldson et al. 2004, Occup Environ Med; Schins, Knaapen 2007,Inhal Toxicol; S. Steiner

Two case studies: Iron-carbohydrates and graphene



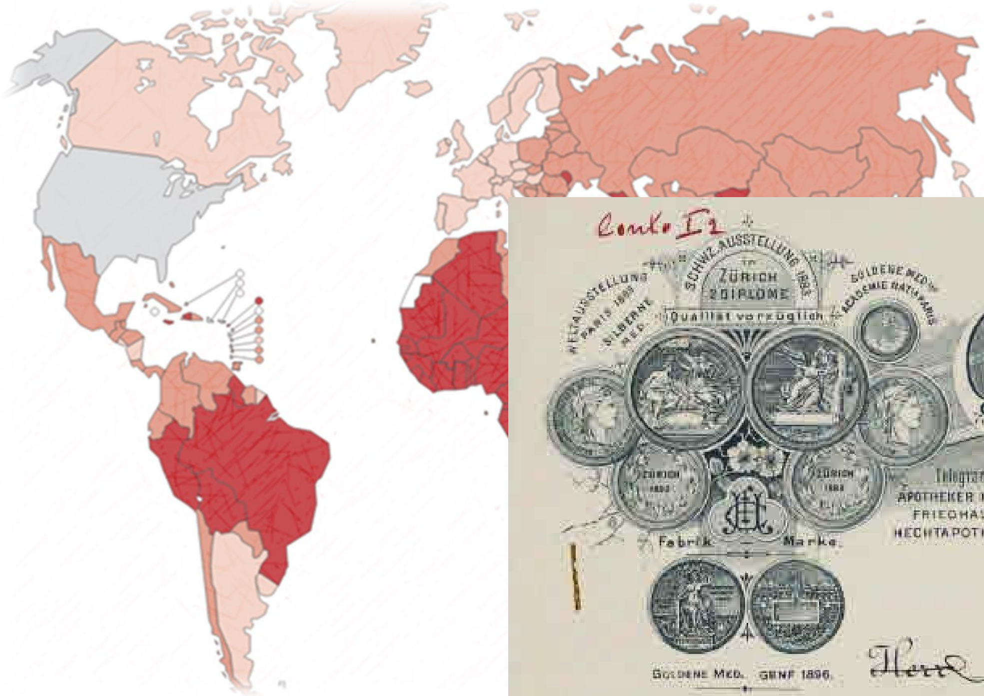


Iron-carbohydrates to treat iron anemia



In collaboration with CSL Vifor

Iron deficiency a world wide challenge



Conto I 2

Beleg No 319

Hecht-Apotheke

Sanitäts-Geschäft

SCHWZ. AUSSTELLUNG ZÜRICH 1883 2 DIPLOME

Qualität vorzüglich

GOLDENE MED.

FRIEDR. HAUSMANN.

Fabrik chemisch-pharmac. Präparate.

Telegramm-Adresse APOTHEKER HAUSMANN, oder FRIEDHAUSMANN, oder HECHTAPOTHEKE

Fabrik Marke

GOLDENE MED. GENF 1896.

St. Gallen, 5. Juli 1897.

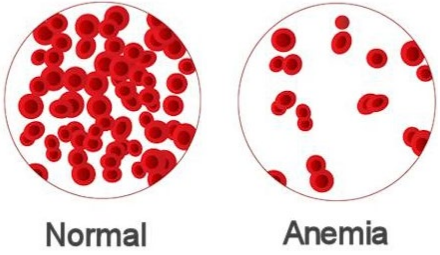
Herrn ... Bauleitung der Klausenstr. Unterschächen

Indem ich Ihnen nachstehend Factura über den mir gütigst ertheilten Auftrag behändige, bitte ich Sie: mich gef. für den Betrag von Fr. 4.75 zu erkennen und mich recht bald wieder mit Ihren werthen Ordres zu erfreuen, deren bester Ausführung Sie sich versichert halten dürfen.

Staatsarchiv St.Gallen

Iron carbohydrate complexes to treat iron deficiency

Iron deficiency (anemia)



Causes:
- Blood loss
- Poor nutrition
- Chronic kidney disease
- Chemotherapy

Role of iron in the body:
- Synthesis of haemoglobin
- Cellular respiration
- Cellular proliferation

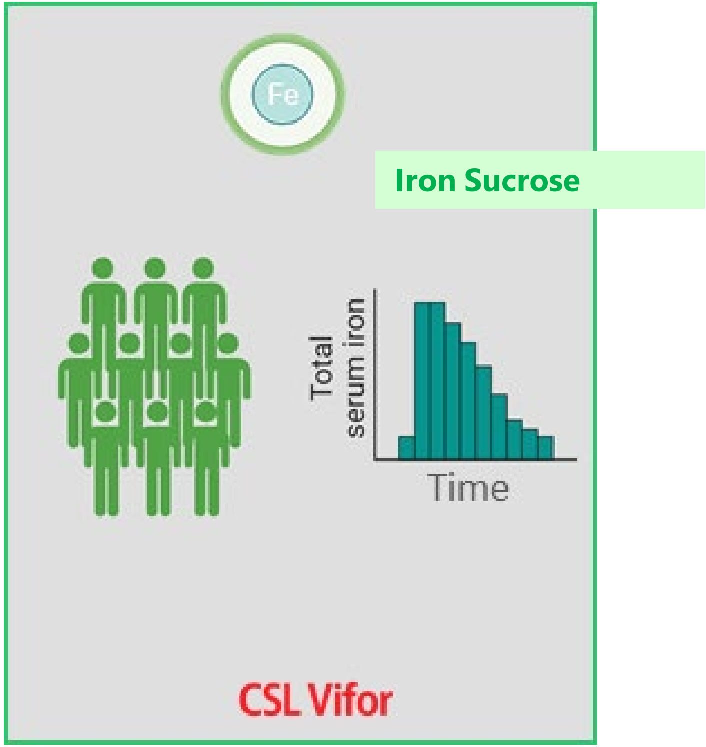
harmful effects on both cells and tissues

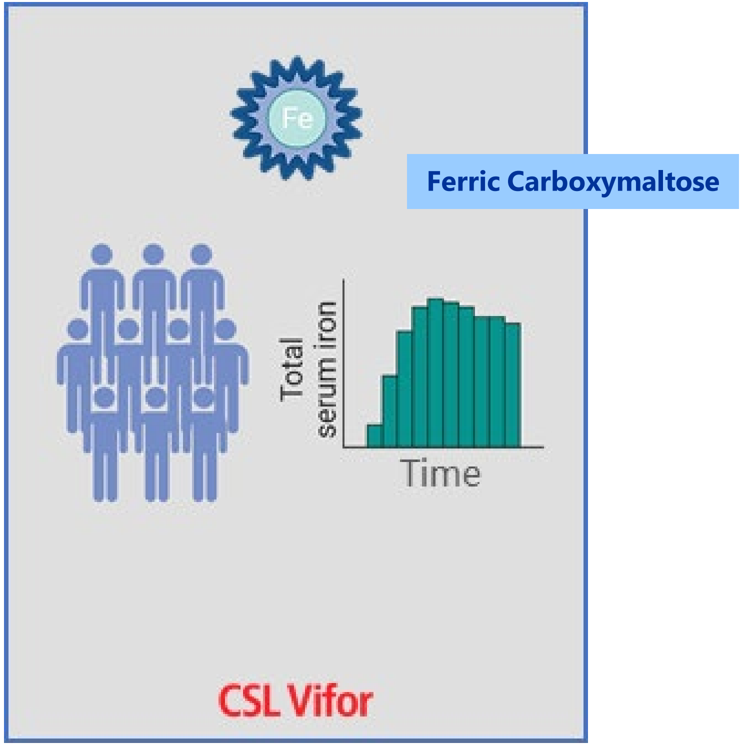
Current treatment

Oral iron supplements

Intravenous iron-carbohydrate complexes

Two Iron-carbohydrate complexes

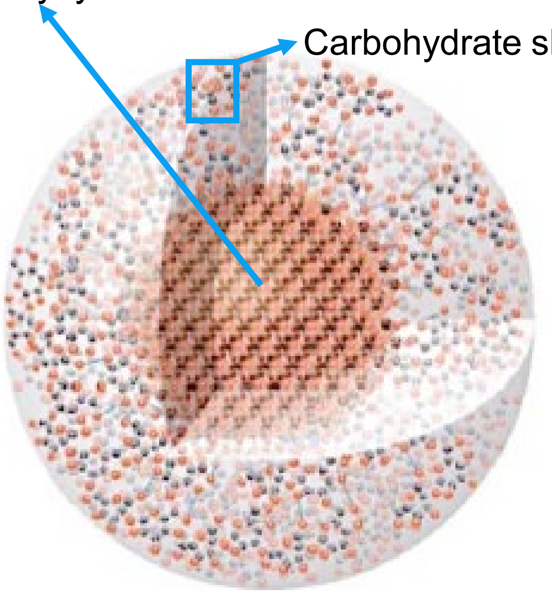




Iron-carbohydrate complexes

Iron (III) oxyhydroxide

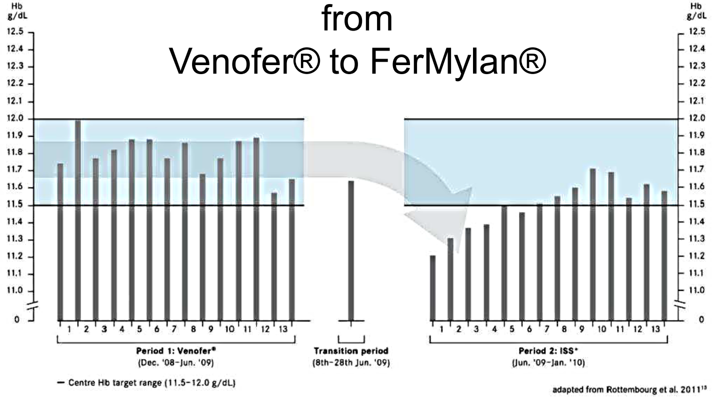
Carbohydrate shell



Iron sucrose
(Venofer, Switzerland)

Intended copies of their originator
(Iron sucrose similar; ISS)

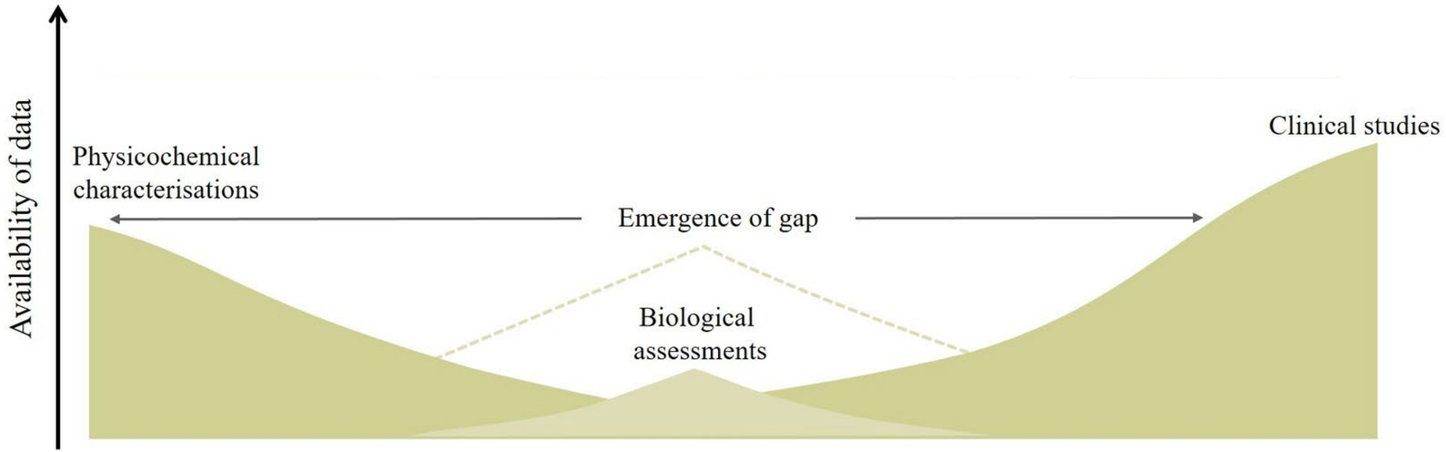
Significant drop in mean Hb after switch from
Venofer® to FerMylan®



N. Nikravesh, et al., 2020, Nanomedicine: Nanotechnology, Biology, and Medicine

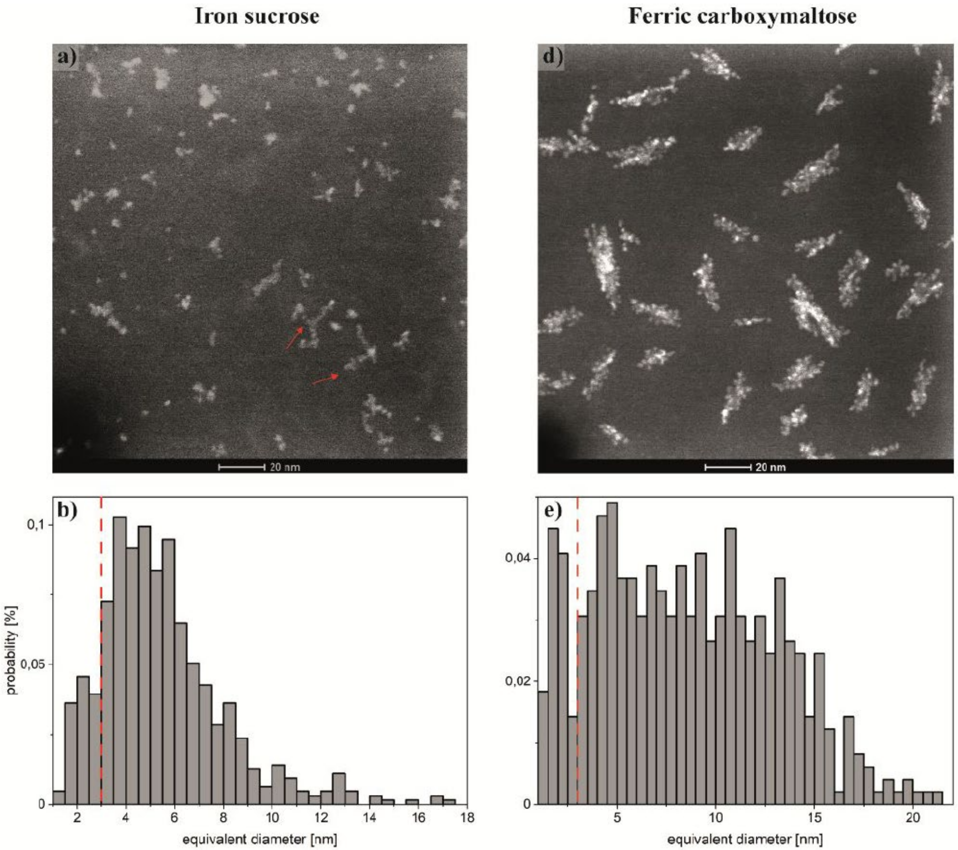
Bridging the gap: Iron-carbohydrate an unusual case

Further **biological assessments** of iron complexes can effectively bridge the gap between pre-clinical and clinical studies.



Nikravesh, N. *et al.* (2020) Nanomed: Nanotech, Bio, and Med

Characterization data Cyro STEM

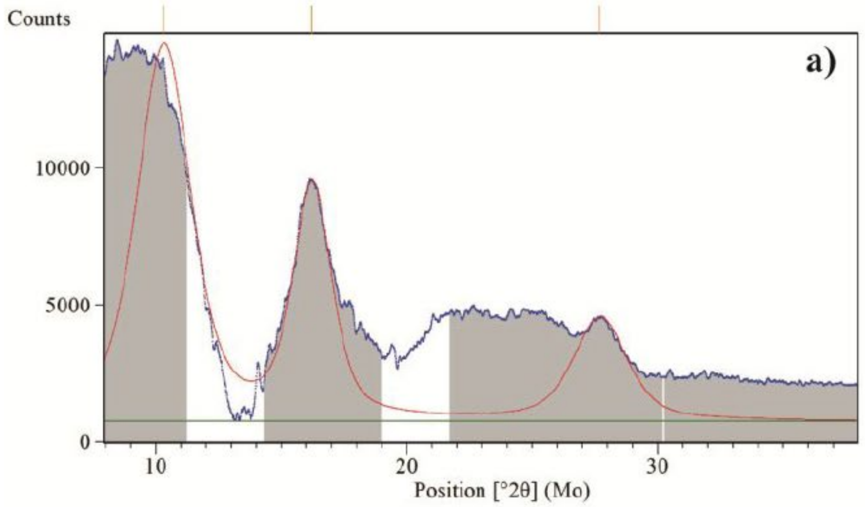


provided by CSL Vifor and ScopeM

morphology and agglomeration

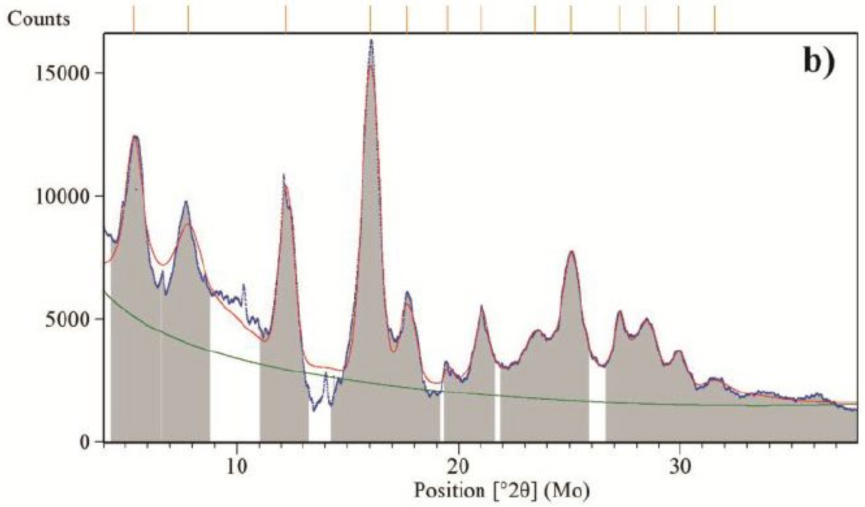
Characterization data of x-ray diffraction (XRD)

Iron sucrose



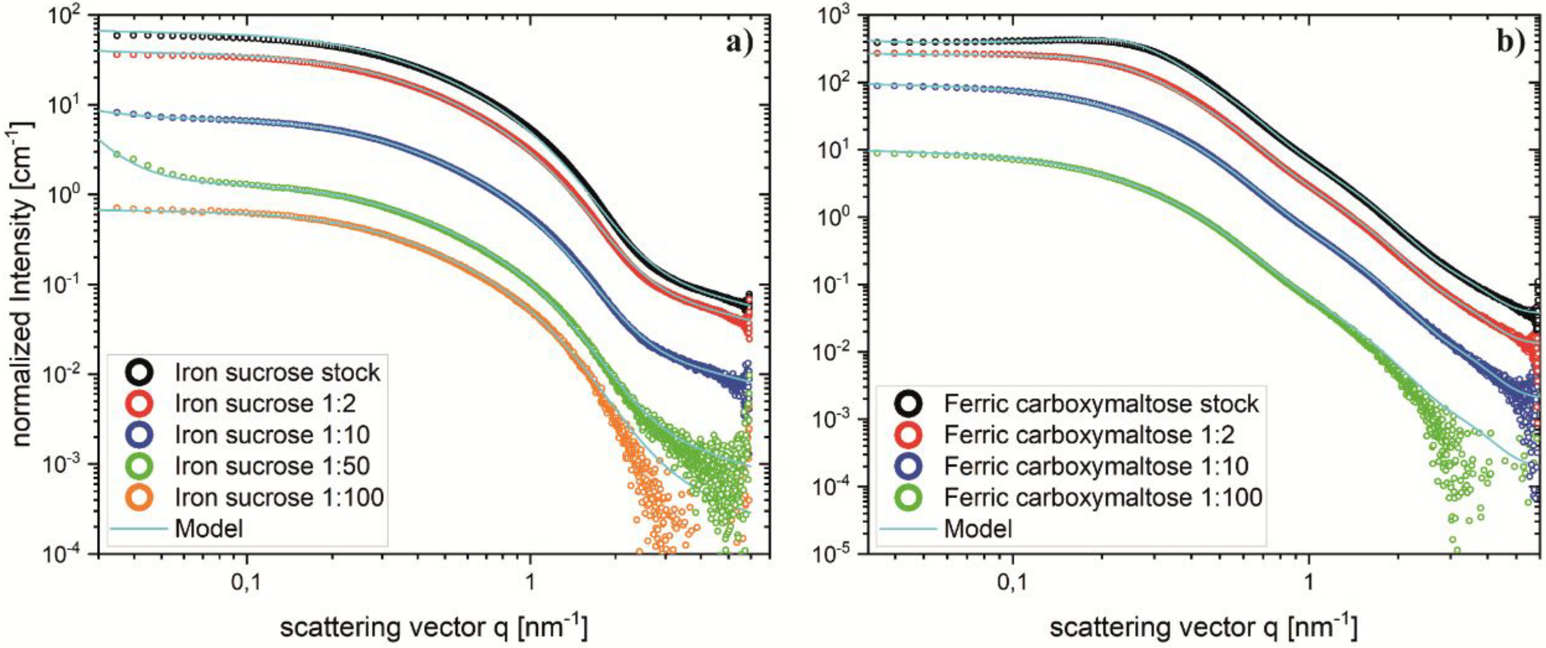
Core: ferroxyhyte crystal structure

Ferric carboymaltose



Core: akageneite crystal structure

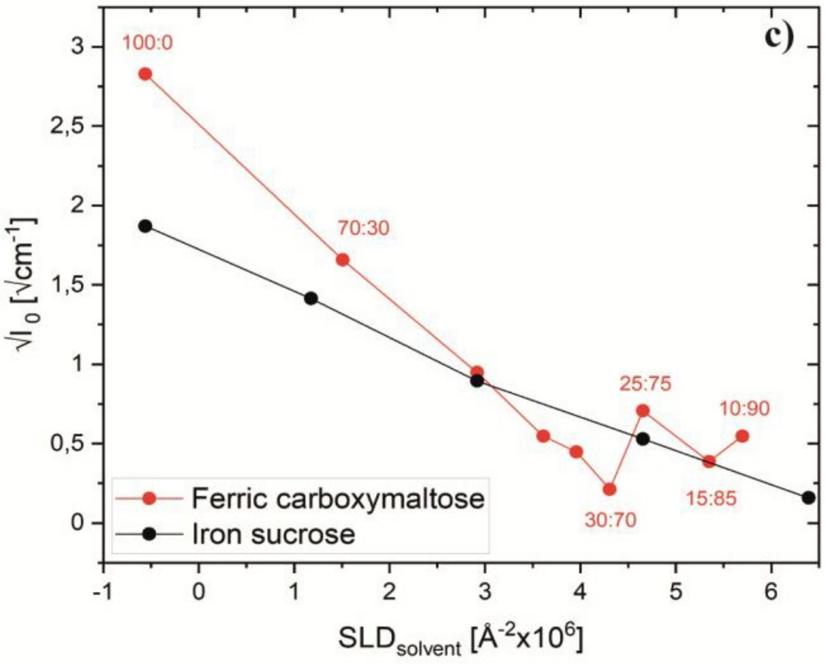
Characterization data from small angle x-ray scattering SAXS

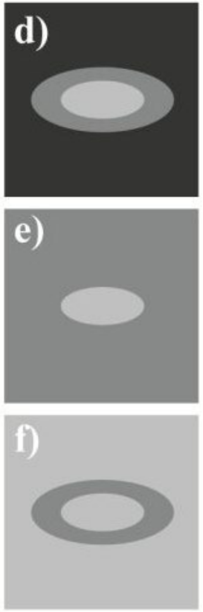


Spheroidal clusters made up of single NPs with unbound sucrose agglomerates

large spheroidal clusters made up of two single NPs with unbound carboxymaltose

Characterization data small angle neutron scattering (SANS)



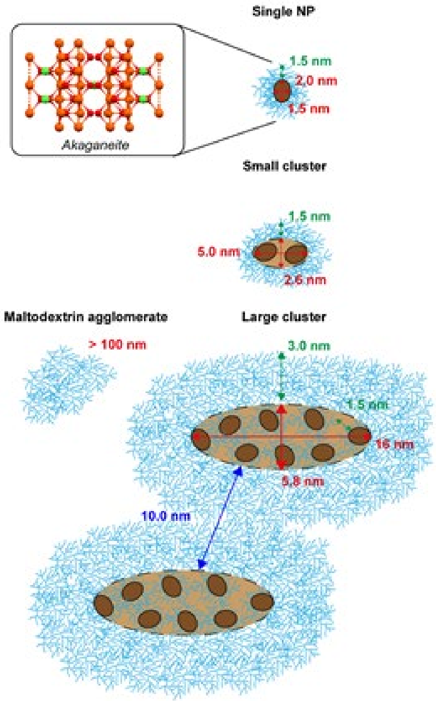


Match point analysis for IS (black) and FCM (red).
(d-f) Schematic representation of a contrast

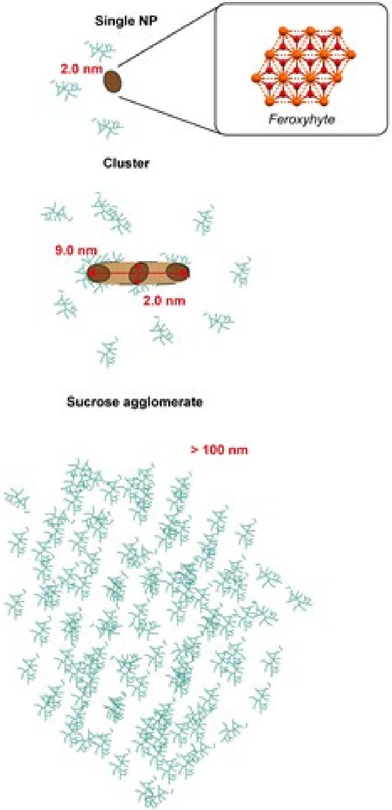
Provide information about shell properties like thinkness

The concluded representation of the nano building blocks and cluster structure

Ferric carboxymaltose



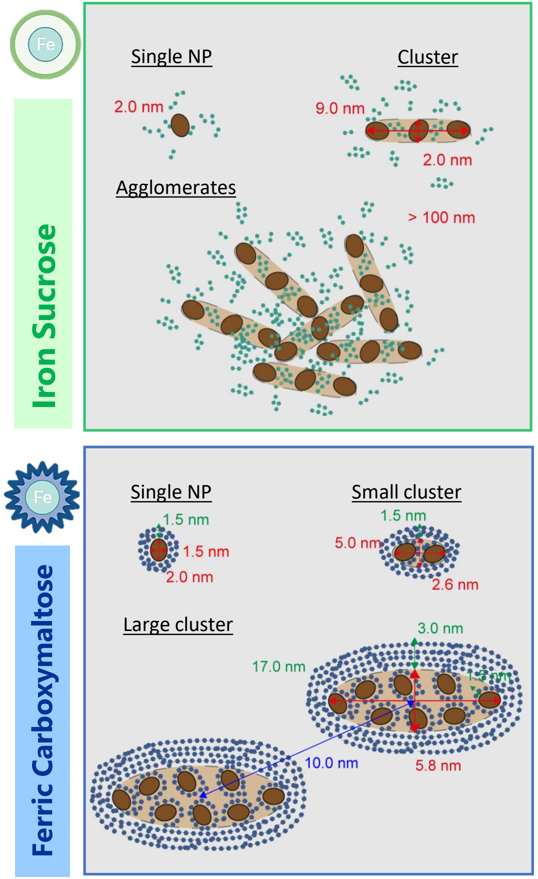
Iron sucrose



Krupnik, Flühmann, Neels, Wick et al 2024 JCR

CSL Vifor

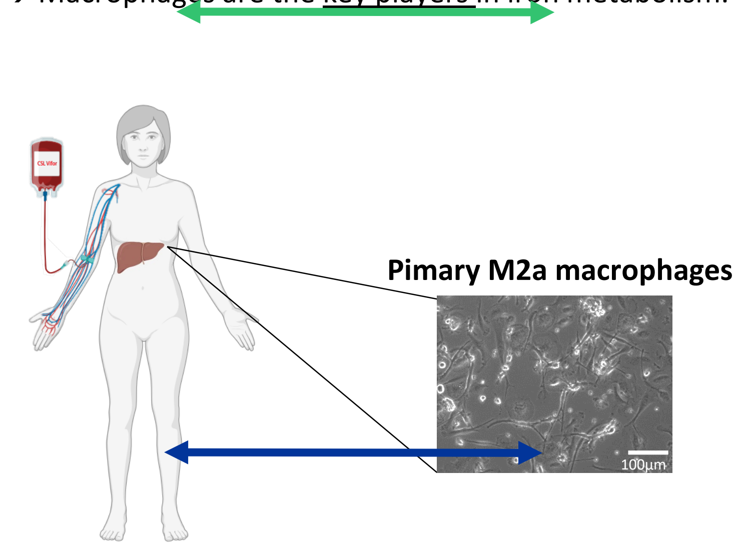
Physicochemical characterization



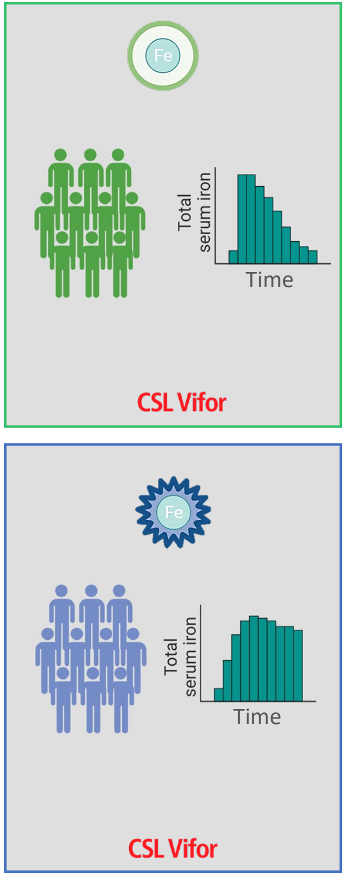
In vitro model

Why Macrophages?

→ Macrophages are the key players in iron metabolism.

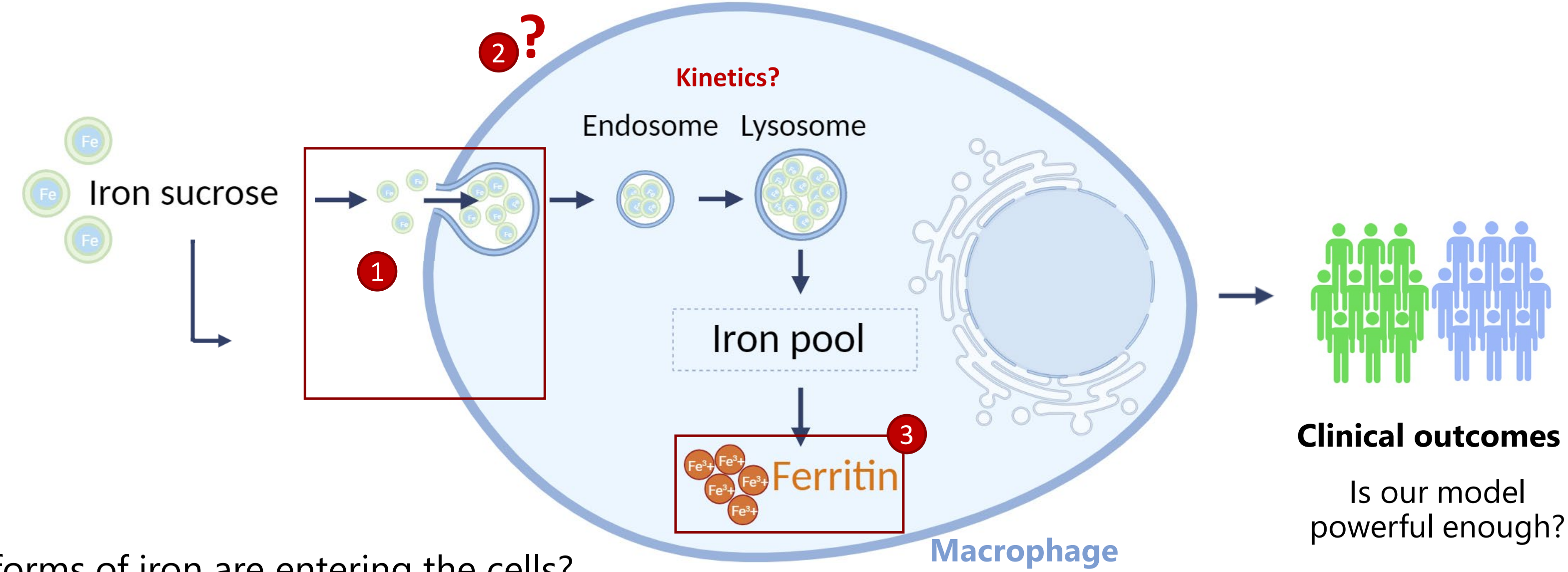


Clinical studies[1]



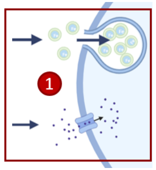
1) Garbowski, M. et al. (2021). Haematologica, 106(11), 2885–2896.

Biological response – State of the art

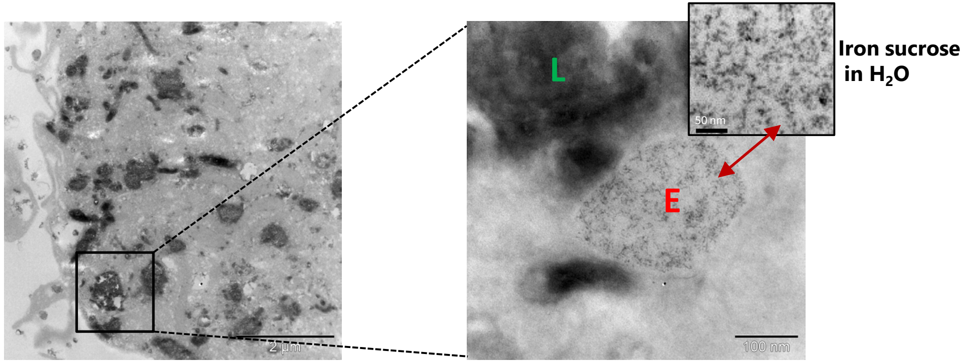


1. Which forms of iron are entering the cells?
2. How does the cell react?
3. Is the iron used and stored?

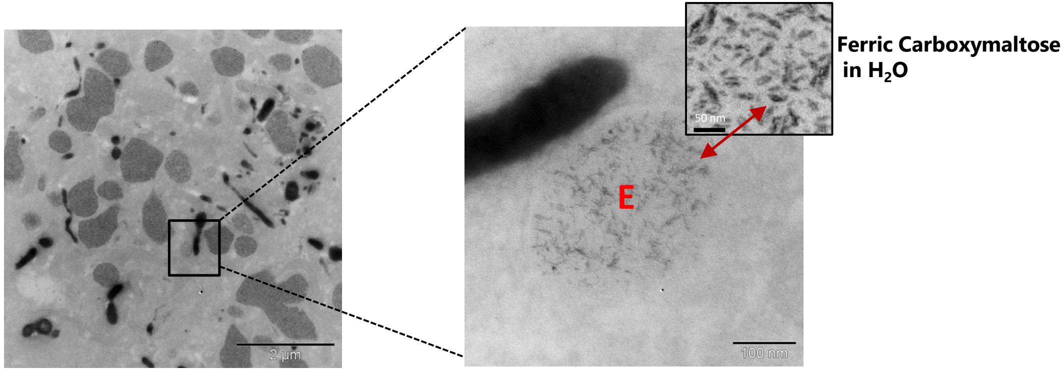
Which forms of iron are entering the cells?



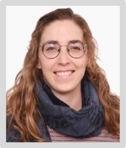
Iron Sucrose



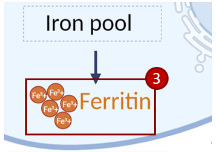
Ferric Carboxymaltose



Complete nanoparticles enter macrophages and may exhibit a therapeutic effect on their own.



Is the iron used and stored in cells?

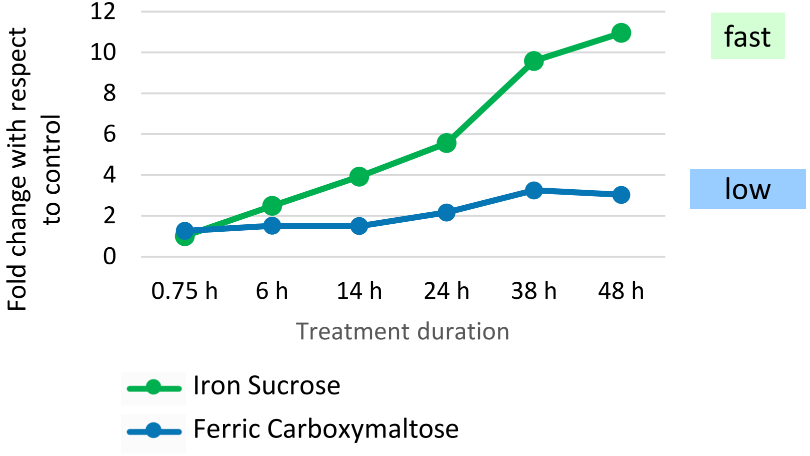


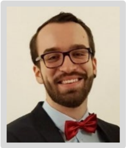
Proteomics[1]

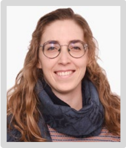
	Iron Sucrose	Ferric Carboxymaltose
Ferritin	3.39	0.89
expression	high	med

$\mathbf{Log_2(FoldChanges)}$

Ferritin production

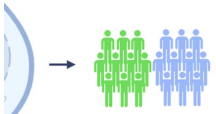






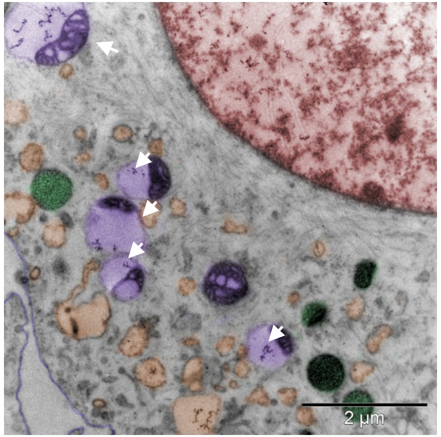
Formulation-dependent variations influence the utilization and storage of iron in macrophages.

Why are there differences in adverse effects?



Iron Sucrose

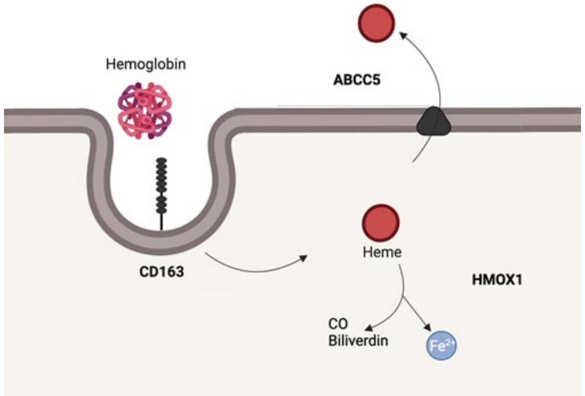
Cells show swollen mitochondria

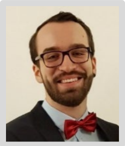


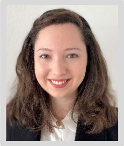
Legend

Mitochondria
Endosomes
Lysosomes
Nucleus

Cells induce mechanisms to prevent iron derived cell death (Ferroptosis)[1]



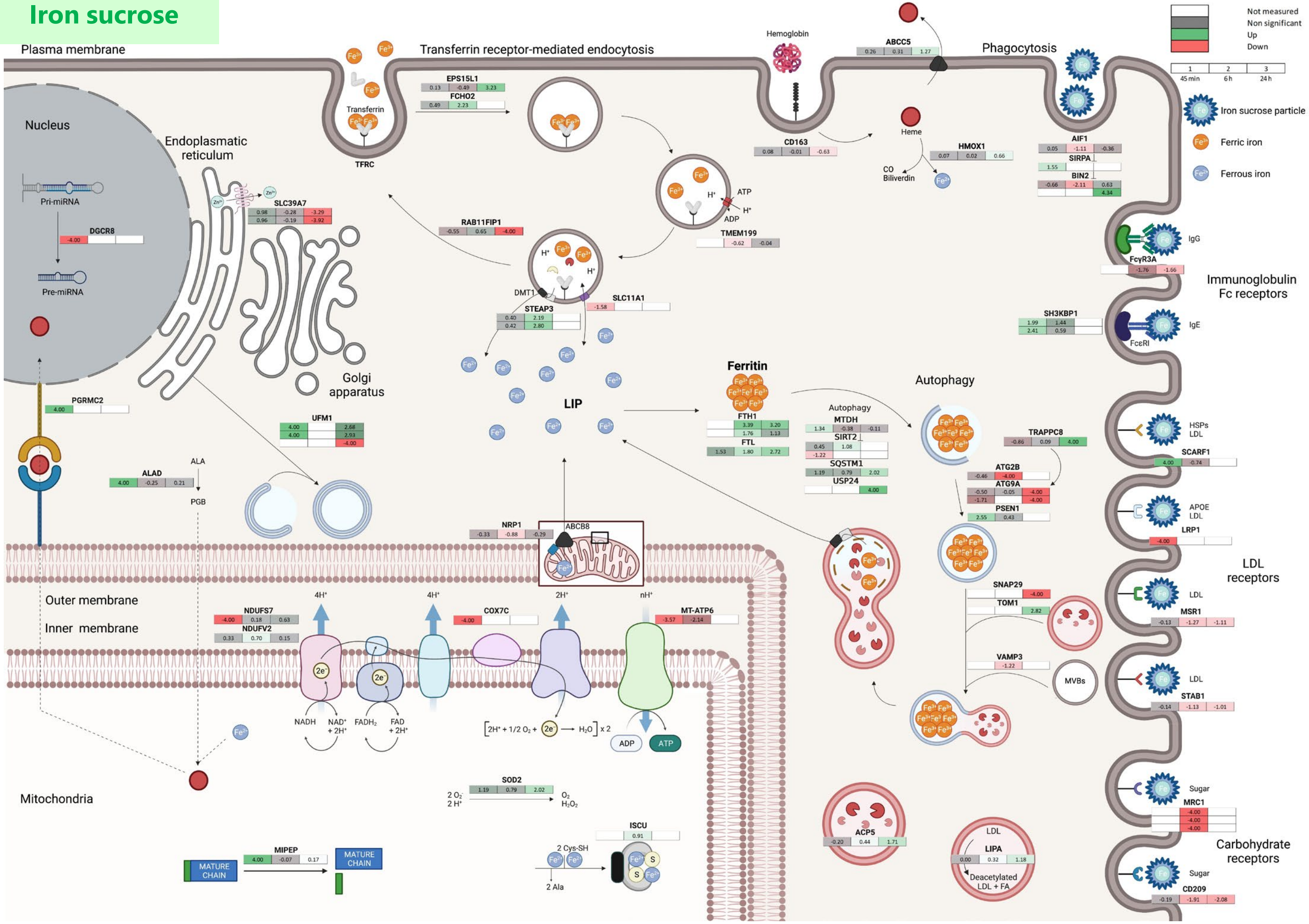


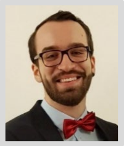


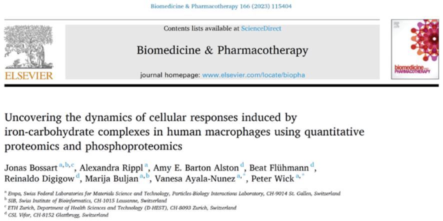
Cytotoxic effects observed at clinical concentrations of IS may contribute to higher adverse effects.

1) Bossart J. et al. (2023) Biomed Pharmacother. 166:115404.

Iron sucrose







Biomedicine & Pharmacotherapy 166 (2023) 115404

Contents lists available at ScienceDirect

Biomedicine & Pharmacotherapy

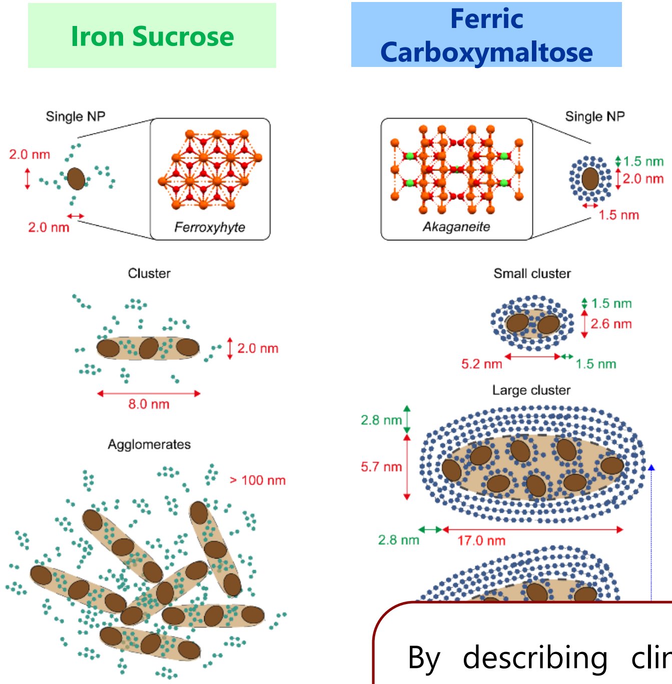
journal homepage: www.elsevier.com/locate/biopha

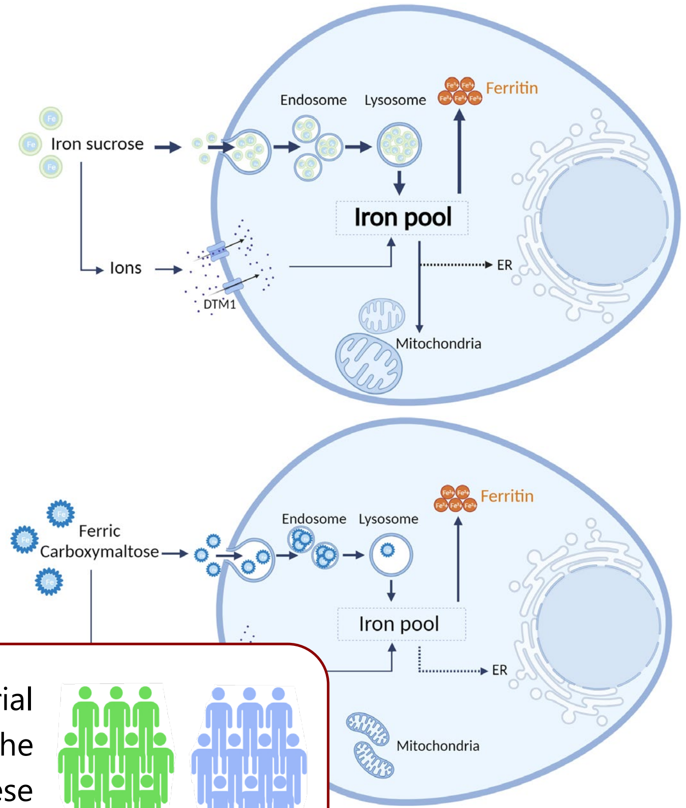
Uncovering the dynamics of cellular responses induced by iron-carbohydrate complexes in human macrophages using quantitative proteomics and phosphoproteomics

Jonas Bossart [a,b,c], Alexandra Rippl [a], Amy E. Barton Alston [d], Beat Flühmann [d], Reinaldo Digigow [d], Marija Buljan [a,b], Vanesa Ayala-Nunez [a,*], Peter Wick [a,*]

[a] Empa, Swiss Federal Laboratories for Materials Science and Technology, Particles-Biology Interactions Laboratory, CH-9014 St. Gallen, Switzerland
[b] SIB, Swiss Institute of Bioinformatics, CH-1015 Lausanne, Switzerland
[c] ETH Zurich, Department of Health Sciences and Technology (D-HEST), CH-8093 Zurich, Switzerland
[d] CSL Vifor, CH-8152 Glattbrugg, Switzerland

Summary and Conclusion

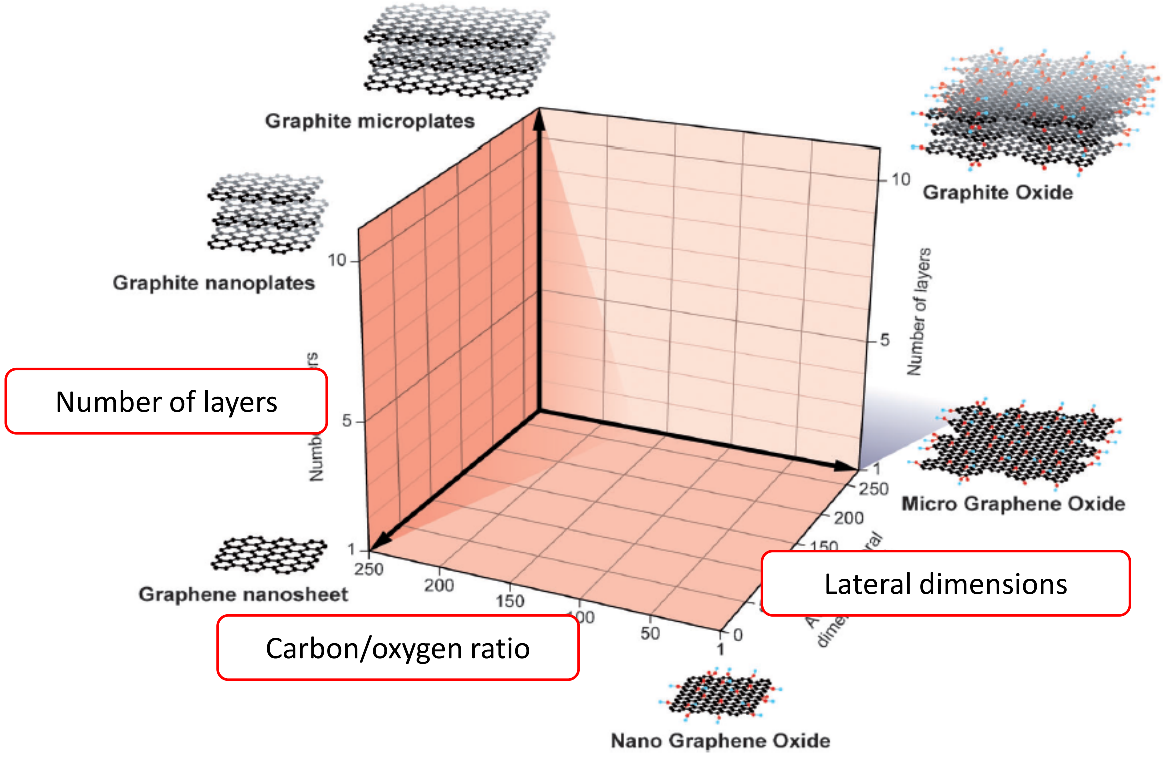




By describing clinically relevant material properties, we now understand the biomedical modes of action of these highquality nanomedicines.

Q&A

Classification framework for graphene related materials

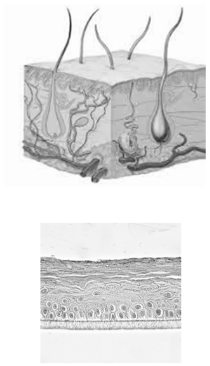


Wick et al. 2014 Angew Chem

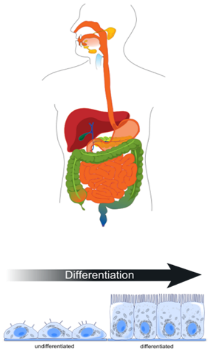
Interaction of graphene related materials with biological barriers and tissues

GRAPHENE FLAGSHIP

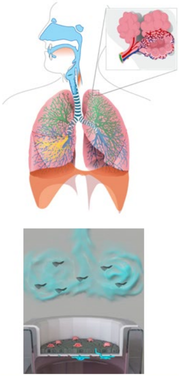
Skin barrier



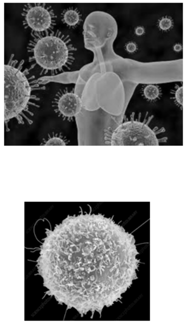
Intestinal barrier



Air-blood barrier

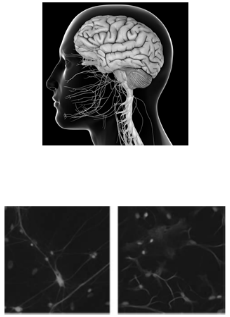


Immune system



Placental barrier

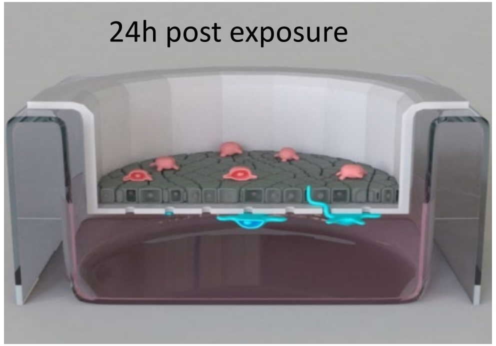
Neuronal system



comprehensive understanding of graphene and graphene related material bio-responses

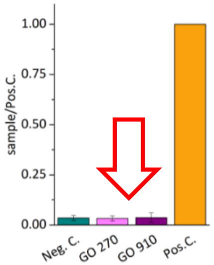
Graphene oxide does not induce an acute tox response in lung *in vitro*

Graphene oxide suspension

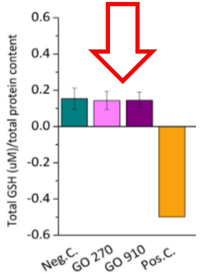


In vitro 3D co-culture

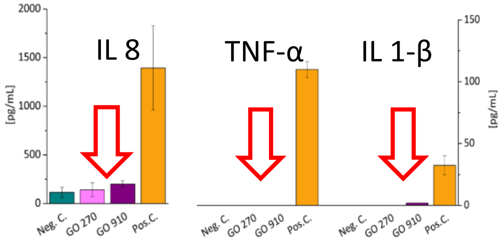
cytotoxicity



oxidative stress



(pro) inflammation



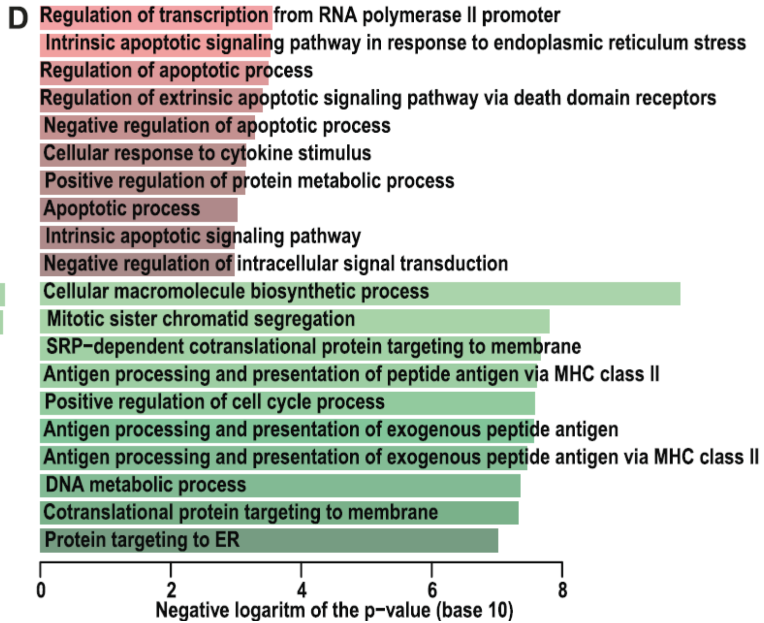
Single exposure to aerosolized GO and GNPs did not initiate an acute biological response in a 3D human lung.

Mukherjee, Wick et al 2016 PLOS One; Drassler, Wick et al 2018 Carbon

Gene expression profiling of human macrophages after GO and GNP treatment induces particle-specific regulation of pathways

MDM 20ug/ml GO 24h

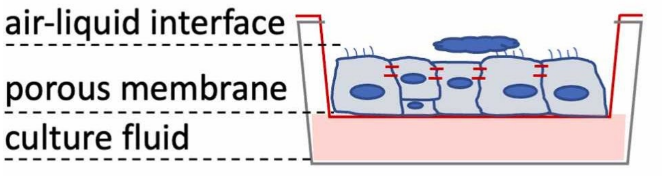
MDM 20ug/ml GNP 24h

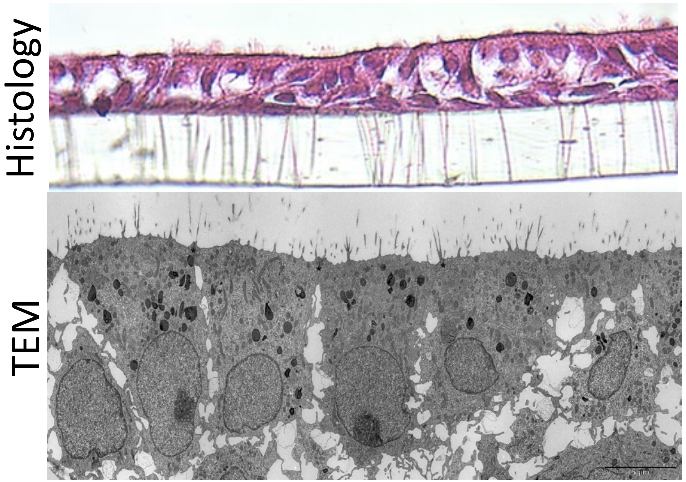


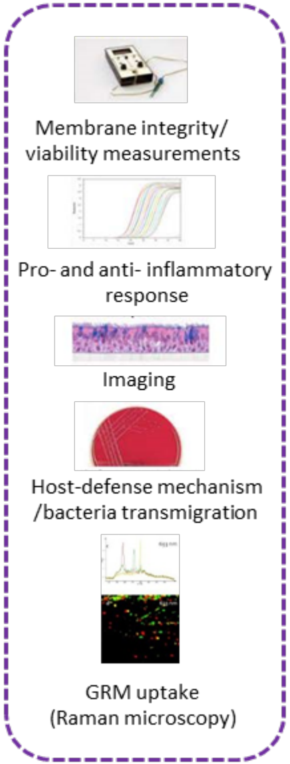
Assessment of long-term and repeated graphene related material exposure on human airway epithelium

In collaboration with:

Kantonsspital St.Gallen







human airway epithelium

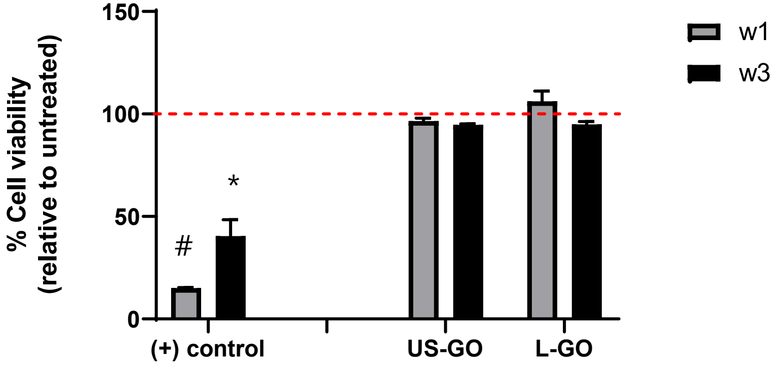
- Primary pseudostratified bronchial epithelium
- Fully differentiated: ciliated, mucus producing and basal cells
- Active mucociliary clearance (MCC)
- Cultured at air-liquid interface conditions (ALI)
- Lifetime and stability of **several months**

GO does not affect cell viability nor barrier integrity

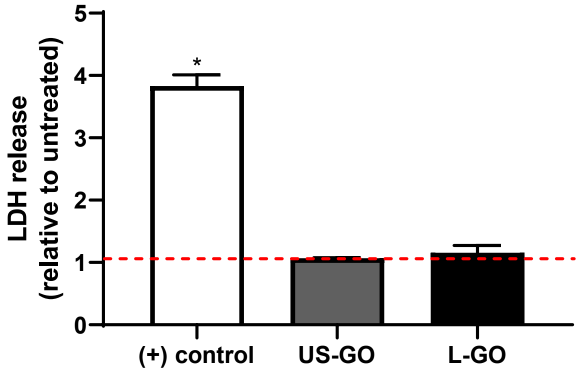
In collaboration with:

Kantonsspital St.Gallen

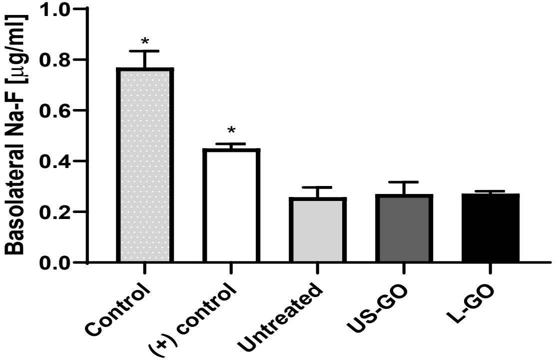
MTS viability assay



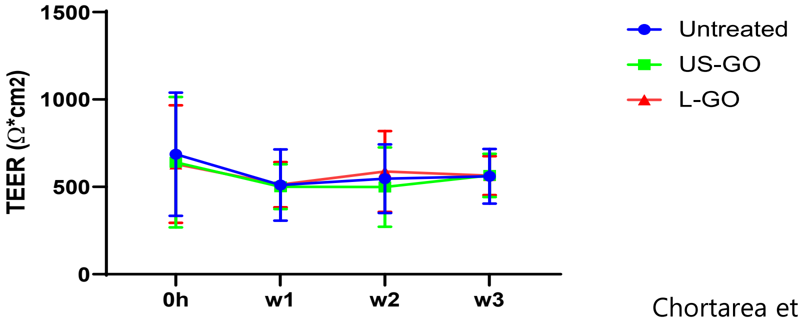
Cytotoxicity



Permeability assay



Membrane Integrity

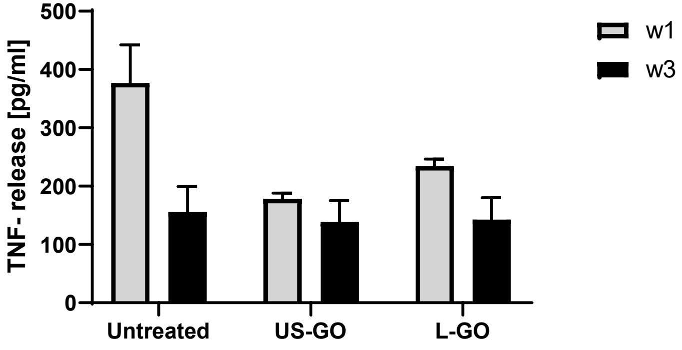


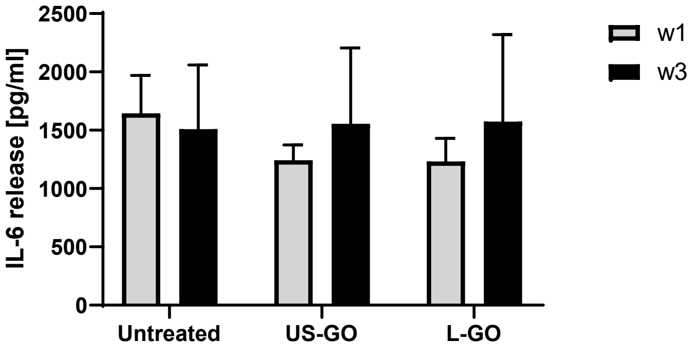
Chortarea et al in prep

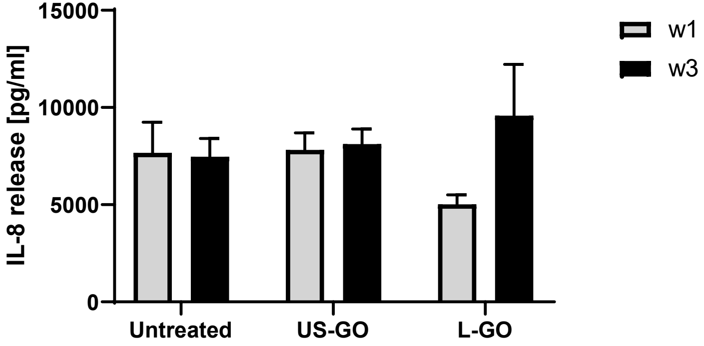
… nor cytokine storming

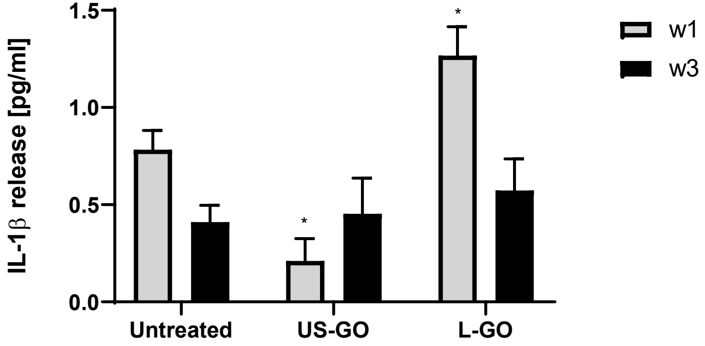
In collaboration with:

Kantonsspital St.Gallen



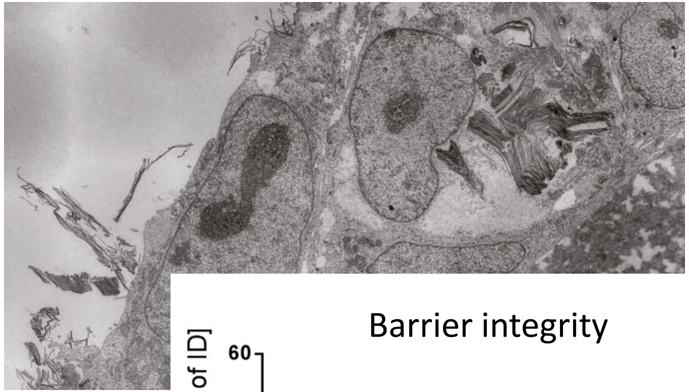


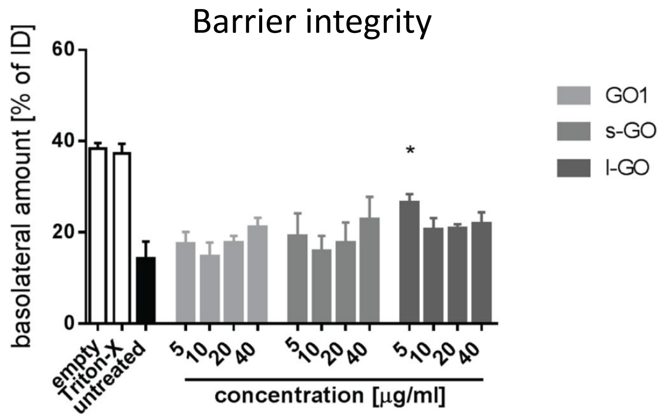




Chortarea et al in prep

Ex vivo human placenta: a unique opportunity to study whole organ – nanomaterial interactions

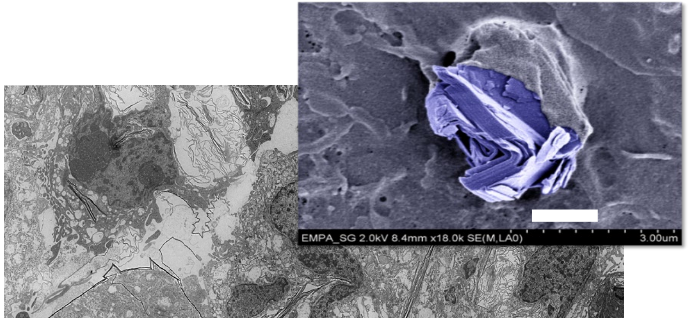




- Consent of the mother
- Placenta is the most species-specific organ in mammalians
- Perfusion duration 6 – 8h, sufficient to calculate the translocation rate
- Provides clinical relevant data

Kucki, Wick et al 2018 2D Materials; Grafmüller, Wick et al 2015 EHP; Wick et al 2010 EHP

Disentangling the structure – activity relationship of graphene and graphene related materials



ACS NANO
Cite This: ACS Nano 2018, 12, 10582–10620
www.acsnano.org

Safety Assessment … Materials: Focus or … Environment

Bengt Fadeel,*† Cyrill Bussy,‡ Sonia M…
Florence Mouchet,|| Lauris Evariste,|| L…
Cristina Martín,# Lucia G. Delogu,¶,§ T…
Didier Beloin-Saint-Pierre,△ Roland His…
Mauro Tretiach,▲ Fabrizia Cesca,▽ F…
Kostas Kostarelos,‡ Maurizio Prato,*○●

ACS NANO
Open Access

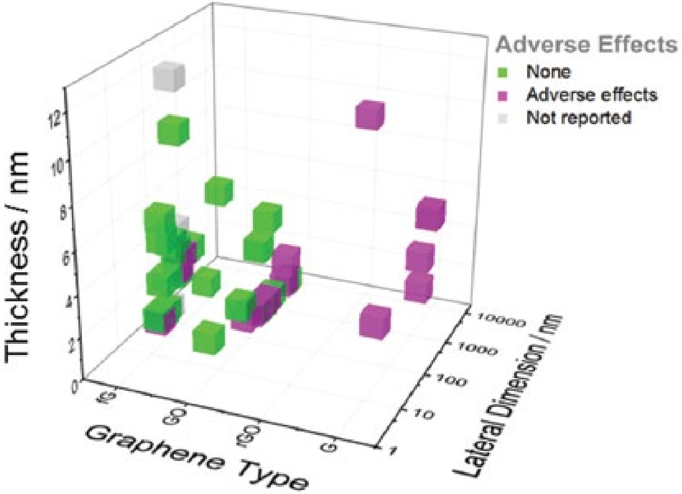
www.acsnano.org

REVIEW

Environmental and Health Impacts of Graphene and Other Two-Dimensional Materials: A Graphene Flagship Perspective

Hazel Lin, Tina Buerki-Thurnherr, Jasreen Kaur, Peter Wick, Marco Pelin, Aurelia Tubaro, Fabio Candotto Carniel, Mauro Tretiach, Emmanuel Flahaut, Daniel Iglesias, Ester Vázquez, Giada Cellot, Laura Ballerini, Valentina Castagnola, Fabio Benfenati, Andrea Armirotti, Antoine Sallustrau, Frédéric Taran, Mathilde Keck, Cyrill Bussy, Sandra Vranic, Kostas Kostarelos, Mona Connolly, José Maria Navas, Florence Mouchet, Laury Gauthier, James Baker, Blanca Suarez-Merino, Tomi Kanerva, Maurizio Prato, Bengt Fadeel,* and Alberto Bianco*

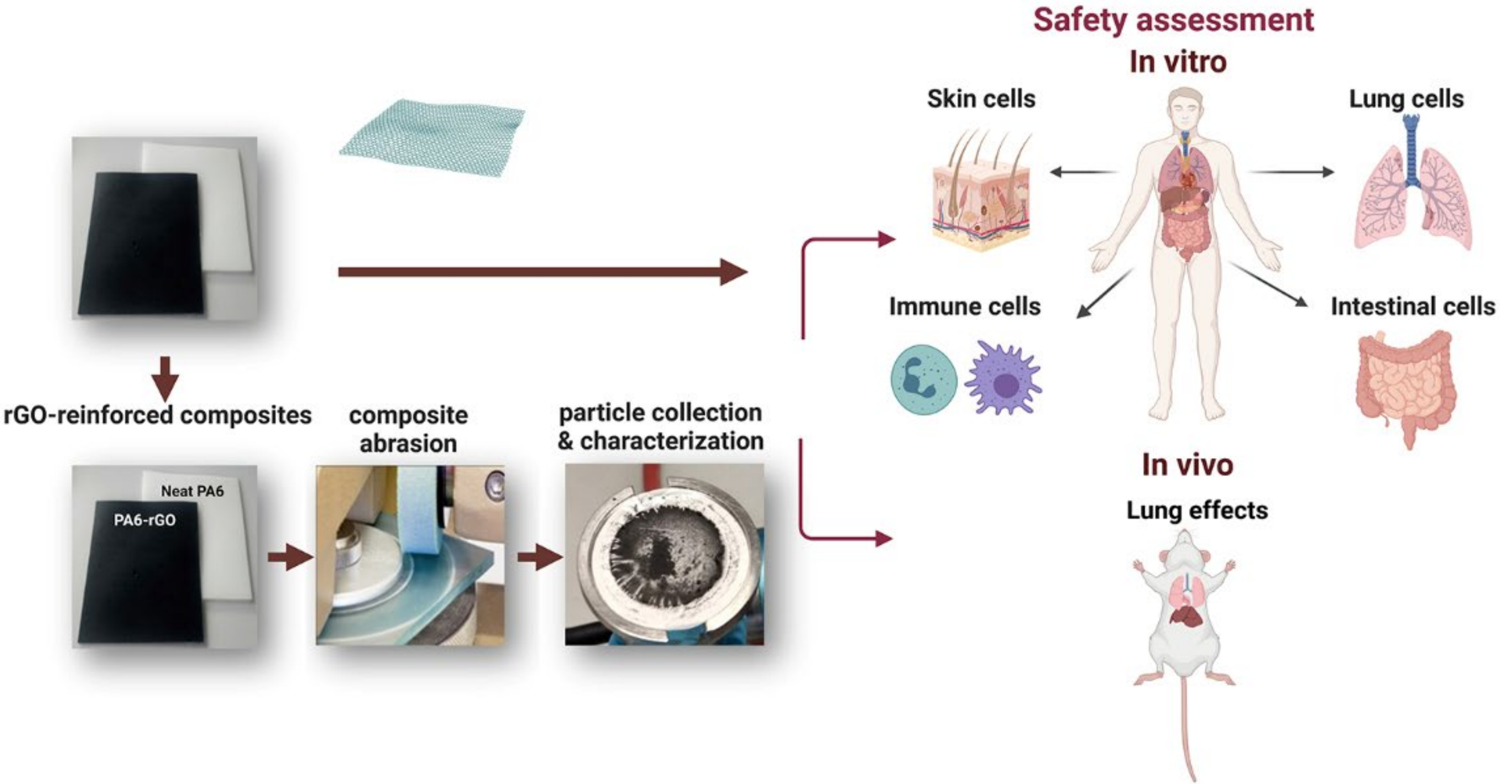
Cite This: https://doi.org/10.1021/acsnano.3c09699
Read Online



Bussy et al. 2015 Nanoscale

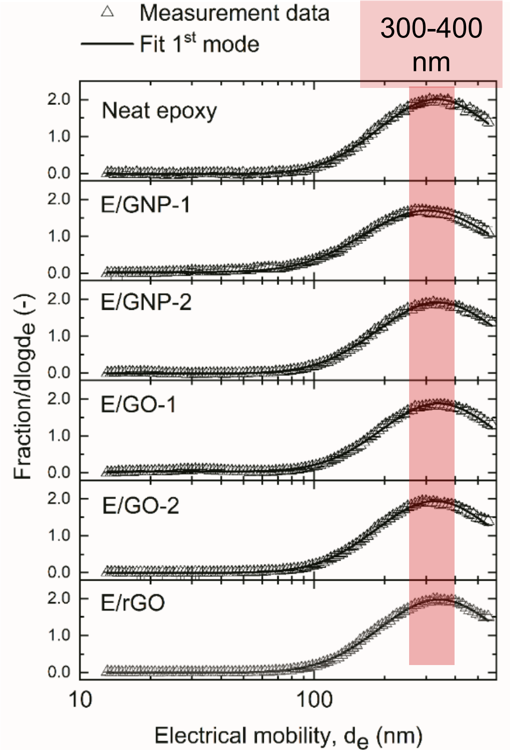
Wick et al 2014 Angew Chem
Kucki et al 2016 NanoScale
Mukherjee, Wick et al 2016 PlosOne
Kucki, Wick et al 2017 J Nanobiotechnology
Kucki, Wick et al 2018 2D Materials
Drasler, Wick et al 2018 Carbon
Fadeel, Wick et al 2018 ACS Nano

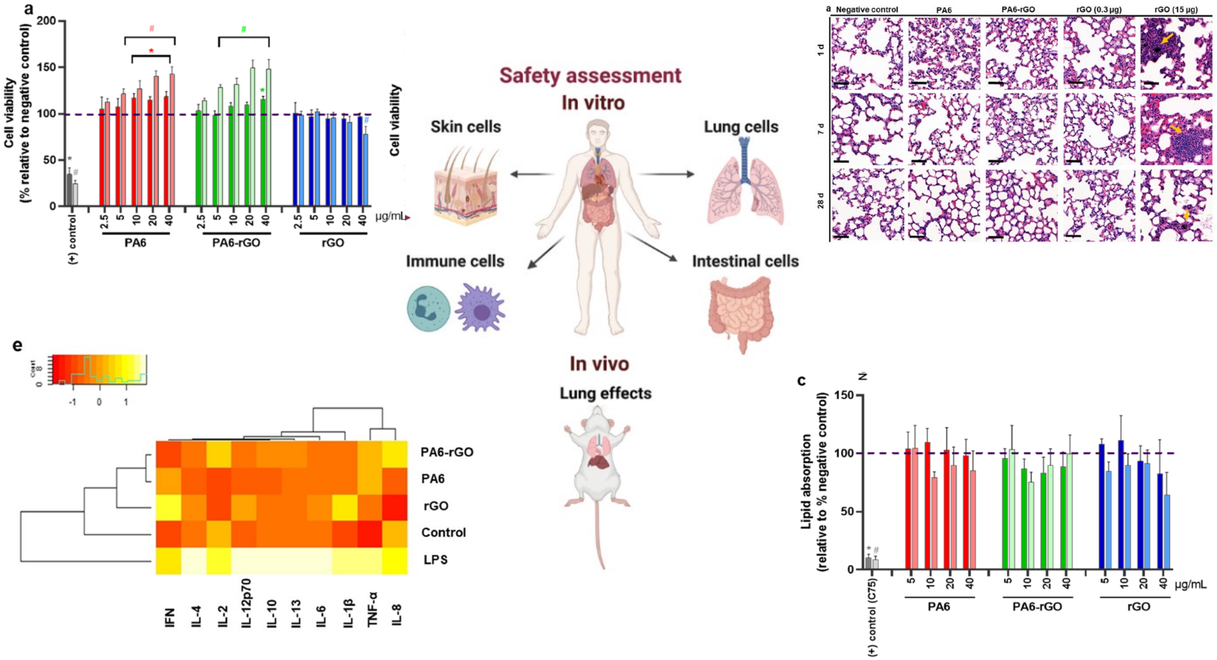
Processed materials and end-of-life assessment of graphene related materials



Chortarea et al J Hazard Mat 2022; Netkueakul et al NanoImpact 2022; Netkueakul et al Nanoscale 2020

Particle size distribution of abraded PA6/GRMs and their potential effect on human health

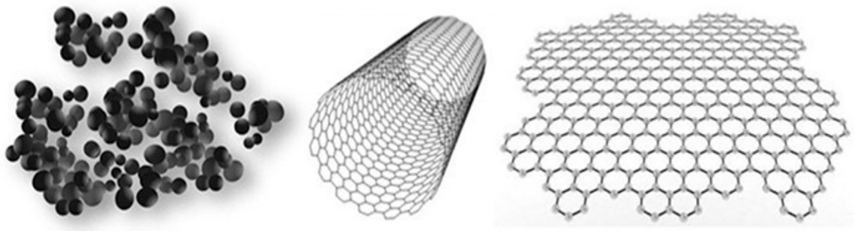




Chortarea et al J Hazard Mat 2022

No significant acute adverse effects of abraded graphene related material-reinforced composites observed

Meta analysis of GRM composites: a release and toxicity assessment in comparison with CB and CNTs



Carbon black — **CB**

Carbon nanotube — **CNT**

Graphene — **GRM**

Data of around 100 studies/publications summarized and evaluated

Romeo, Clement, Wick under revision

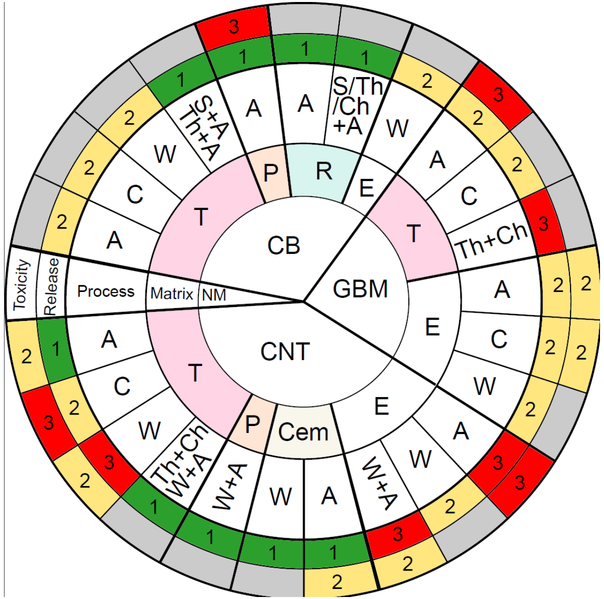
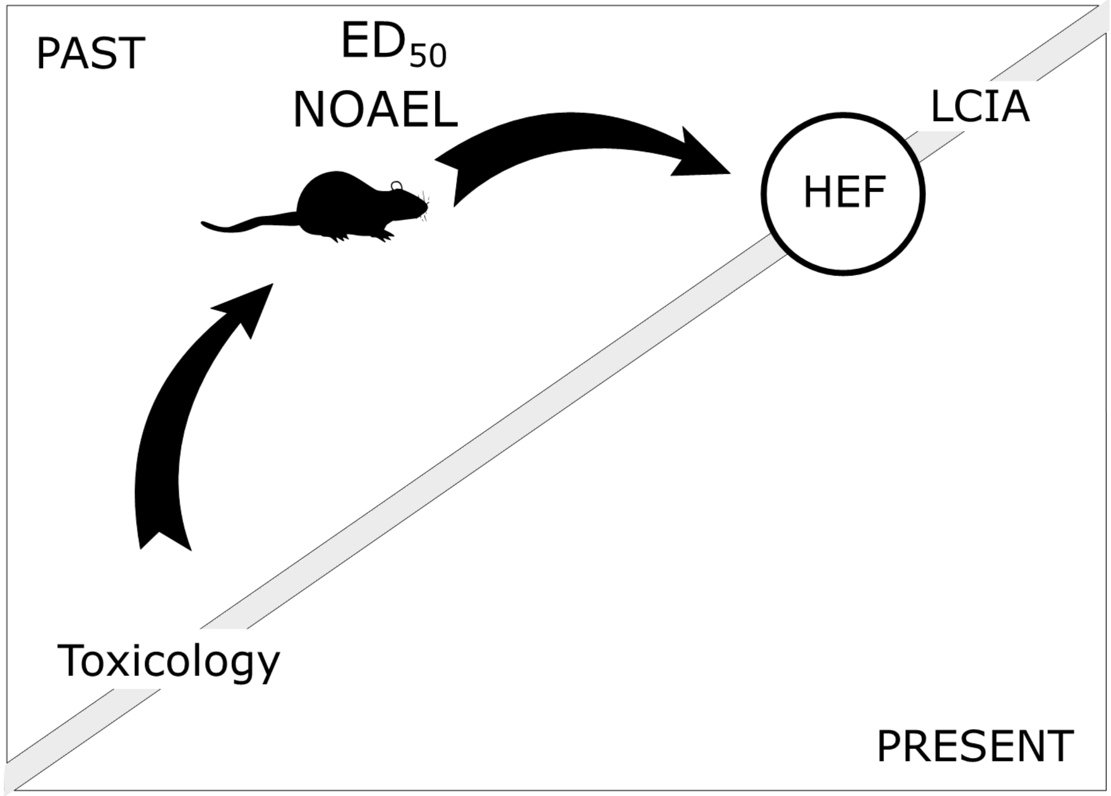


Figure 5: Overview of the classification of release and toxicity information (as of July 2022). For the matrix: T= thermoplastics; P= paints; R= rubber; E= epoxy; Cem= cement. For the processes: A= abrasion; C= combustion; W= weathering; S= stretching; Th= thermal stress; Ch= chemical stress. Green (1): no free NM released (for release) or no nano-specific effect verified *in vivo* or with multiple *in vitro* tests (for toxicity). Yellow (2): Free NM detected only qualitatively or not investigated (for release), or too few studies showing no nano-effect (for toxicity). Red (3): free NM detected with a quantitative method (for release) or additional nano-effect observed (for toxicity), Grey: no data available

How to make *in vitro* data useful for Life Cycle Impact Assessment (LCIA) and Risk Assessment (RA)

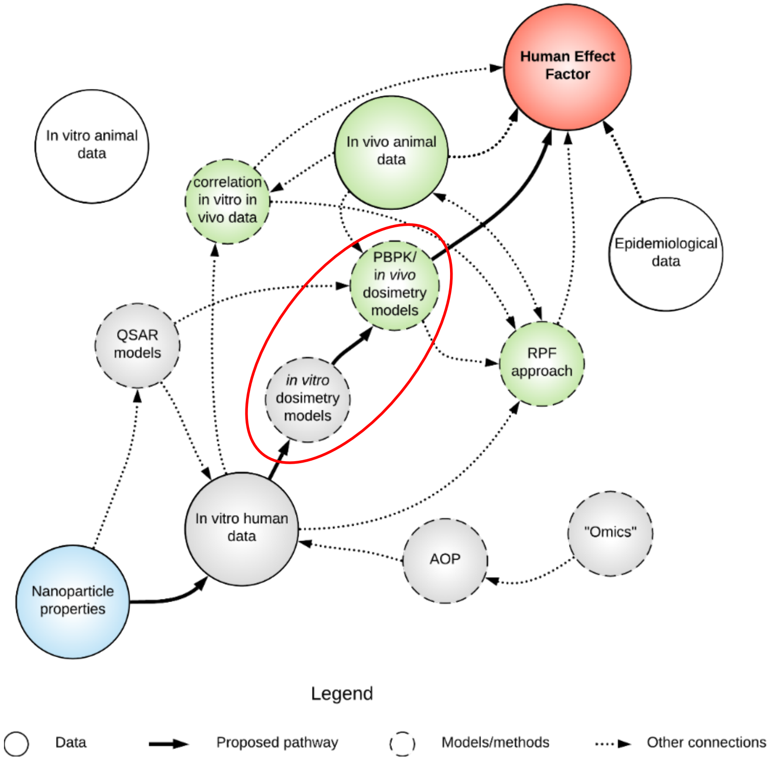


How to use *in vitro* data in place of animal studies in assessment methodologies such as Risk Assessment (RA) and Life Cycle Impact Assessment (LCIA)?

HEF = Human effect factor
Indicates the incidence of a disease caused by the intake of a chemical or other substance

ED = effect dose
NOAEL = no adverse effect level

An integrated pathway based on in vitro data for the human hazard assessment of nanomaterials

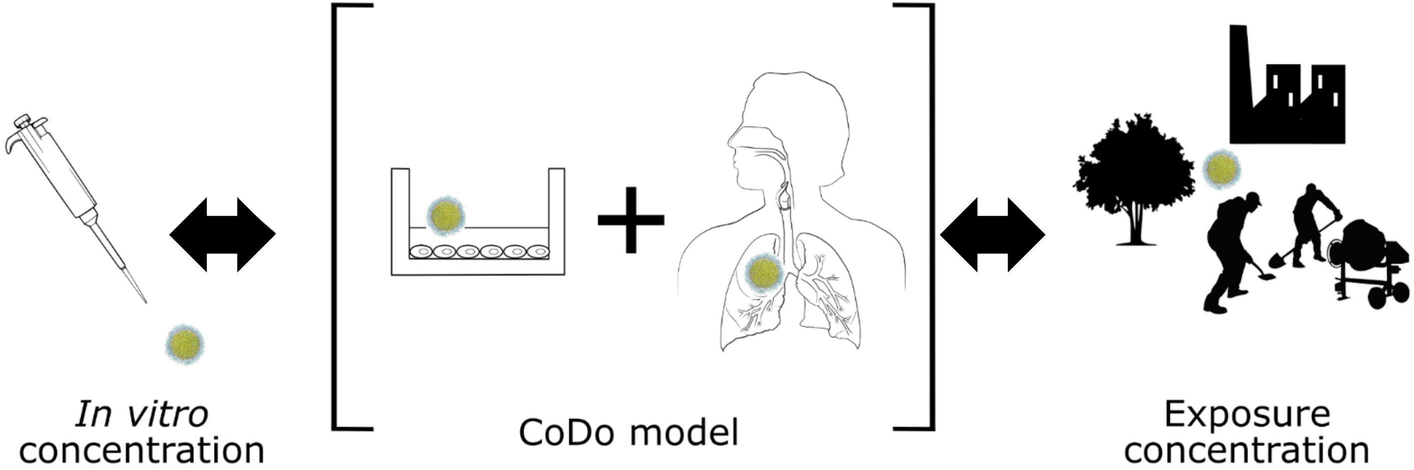


Mapping the existing landscape of models and methods and their potential integration

Strategy chosen to have a wide coverage of nanomaterials and to be testable with available literature data

Romeo, D et al 2020 Environment International
Romeo, D et al 2022 Environ Science Nano

CoDo model: a streamlined approach to in-vitro-to-in-vivo extrapolation

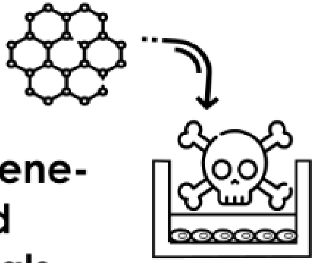


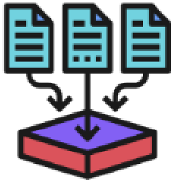
(Model available at https://doi.org/10.5281/zenodo.4889169)

Romeo, D., Nowack, B., & Wick, P. 2022 *NanoImpact*
Romeo, D., Hischier, R., Nowack, B., Wick, P., *2022 EST*

Machine learning approaches for Risk Assessment and Life Cycle Impact Assessment GRMs

DATA COLLECTION	ANALYSES	RESULTS
Graphene-related materials toxicity on lung cells *in vitro*	**Machine Learning** Viability prediction via **regression models** BMDL prediction via regression and **classification models**	**R^2=0.77 Viability prediction** **75% Accuracy BMDL range prediction** Experimental conditions are important



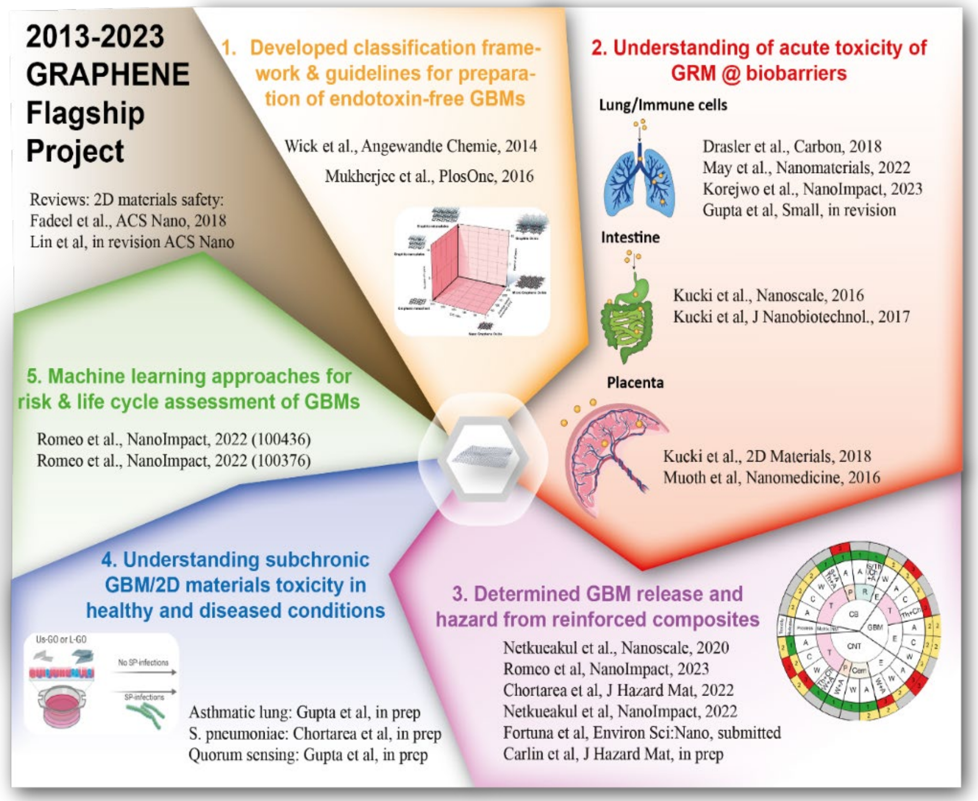


BDML = Benchmark Dose Level

Romeo, Wick et al. NanoImpact 2022

Summary and conclusions

- The life cycle of the nanomaterial-enabled products determines the risk scenario
- Graphene related materials are an excellent setting for studying structure-activity relationships prerequisite for predictive toxicology
- Holistic hazard assessment from freshly manufactured GRMs to abraded GRM-enabled composites revealed no or low acute toxicity at different bio-barriers
- Exploring the safety of other 2D materials such hBN, Graphene acid, Mxenes, etc
- High quality data for future data processing needed for RA and LCIA
- Exploring the potential of ML and big data analysis for GRM safety assessment



… and you for your attention

Collaborations

Prof. S. Sturla, Prof. S. Schürle and teams D-HEST ETHZ
Prof. H. Hofmann, Prof. F. Stellacci and teams EPFL
Prof. G. Borchard, Uni Genf
Prof. J. Huwyler, Prof S. McNeil and team UniBasel
Prof B. Rothen-Ruthishauser, Prof. A. Fink and team AMI
Prof W. Jochum, Prof. R. Hornung, PD M. Neidert KSSG
Prof. Dr. Antonia Neels Empa / University of Fribourg
Prof B. Fadeel and team KI
Prof A. Bianco, Prof M. Prato Graphene Flagship
Prof K Kostaleros and team Uni Manchaster
Prof R. Zboril VSB Technical University of Ostrava, Czech Republic
Dr. R. Schmid SINTEF
Dr. P. Boiseau CEA Grenoble
Dr L. Tran IOM
Prof B. Yan Shandong University, Jinan China
Dr. J. Elliot and team NIST
Prof. K Savolinen, Prof. D.Grecco and teams FIOH
Dr N.W. Song and team KRISS
Dr. Beat Flühmann, Dr. Amy E. Barton Alston,
Dr. Eric Philipp CSL Vifor

GRAPHENE FLAGSHIP

Swiss National Science Foundation

NOVA

metrino

MACRAMÉ

TOXBOX

CHIASMA

INSIGHT

CSL Vifor