Aquatic ecotoxicity testing of nanoplastics

lessons learned from nanoecotoxicology

Hartmann, Nanna Isabella Bloch; Nolte, Tom; A. Sørensen, Mads; R. Jensen, Pernille; Baun, Anders

Publication date:
2015

Document Version
Publisher's PDF, also known as Version of record

Technical University of Denmark

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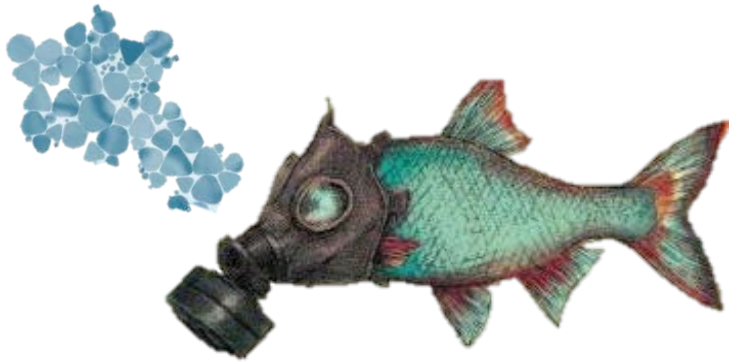
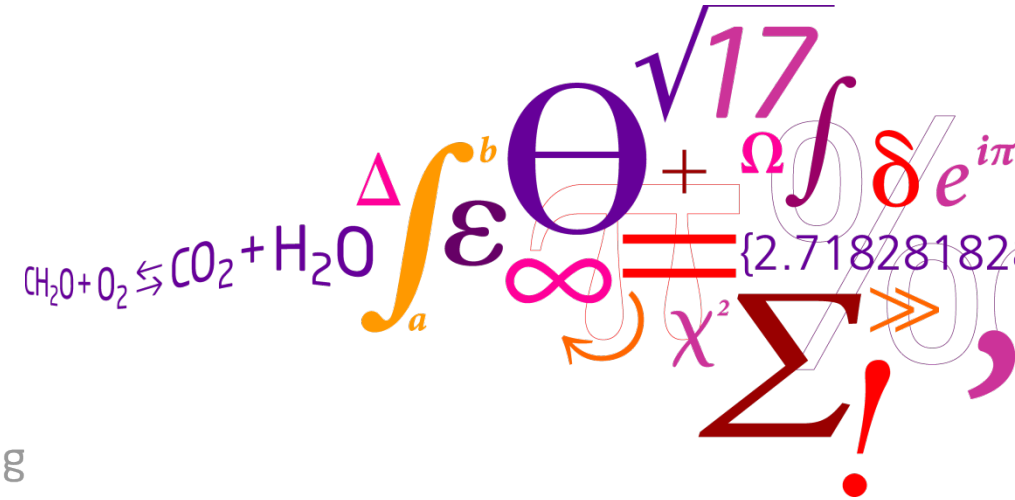


Nanna B. Hartmann, Tom Nolte, Mads A. Sørensen, Pernille R. Jensen, Anders Baun

2015 Aquatic Sciences Meeting
27th February 2015, Granada, Spain



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The group

- Professor Anders Baun
- Associate professor Steffen Foss Hansen
- Senior Researcher
  Hans-Christian Holten Lützhøft
- Postdoc Nanna Bloch Hartmann

- PhD students:
  - Sara Nørgaard Sørensen
  - Denisa Cupi
  - Lars Michael Skjolding
  - Biase Liguri
  - Katrine Nielsen
  - Laura Heggelund
  - Aiga Mackevica
  - Rune Hjort

What do we do?

Hazard Identification

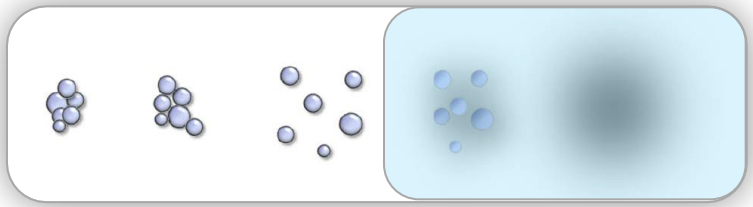
NanoREM

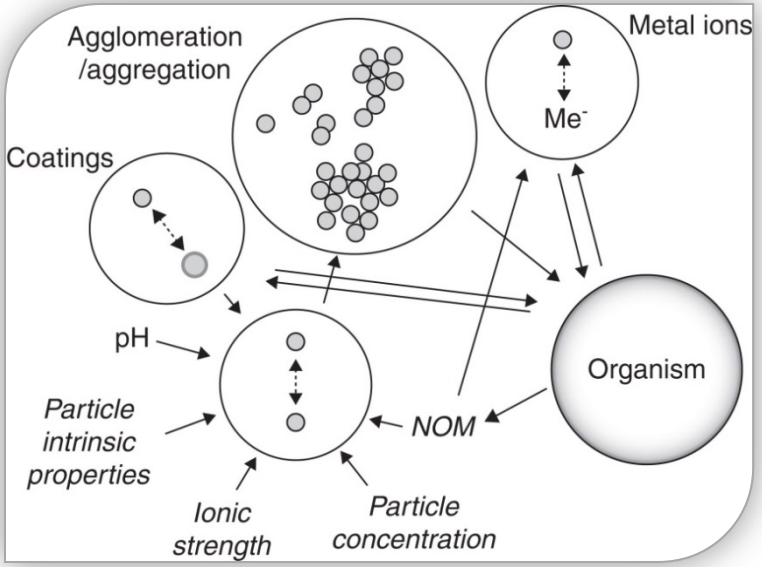
MARINA

NanoDen

EnvNano

New paradigm for ecotoxicity testing of NMs (European Research Council Starting Grant)





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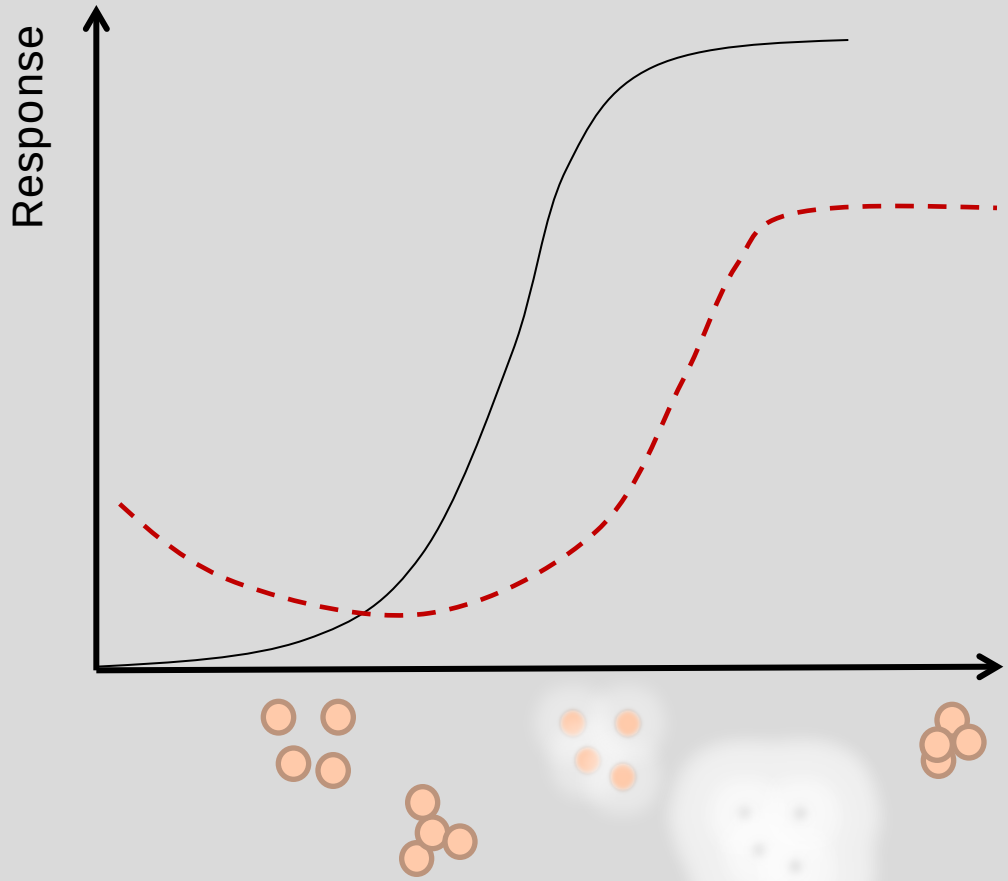
Dose-response testing with nanoparticles

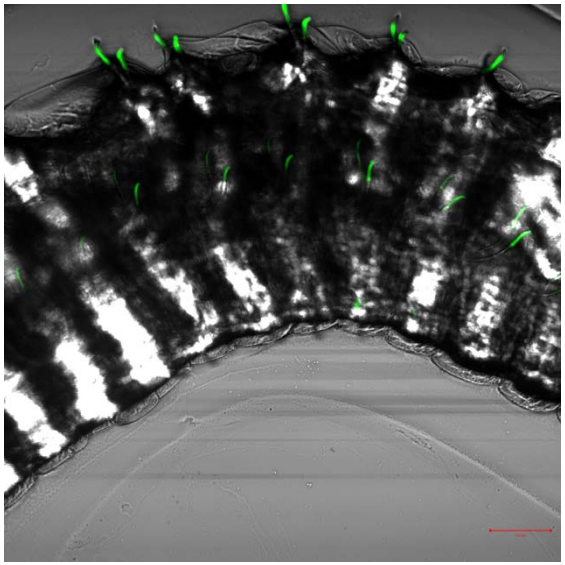


Dose?

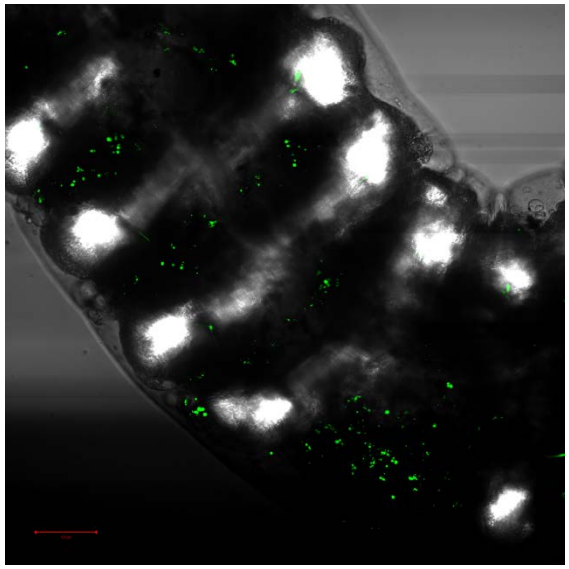
Tuning the test system…

Plastic particles as model particles…

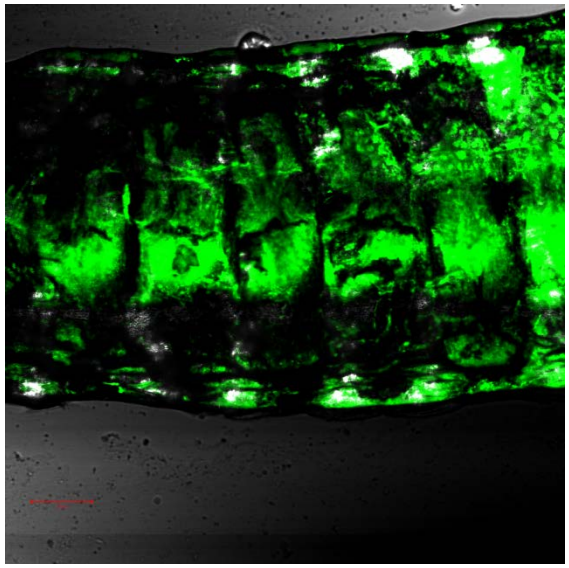
Lumbriculus variegatus (freshwater sediment worm). Aquatic exposure to micro- and nanoplastics. Imaged by confocal microscopy



Control

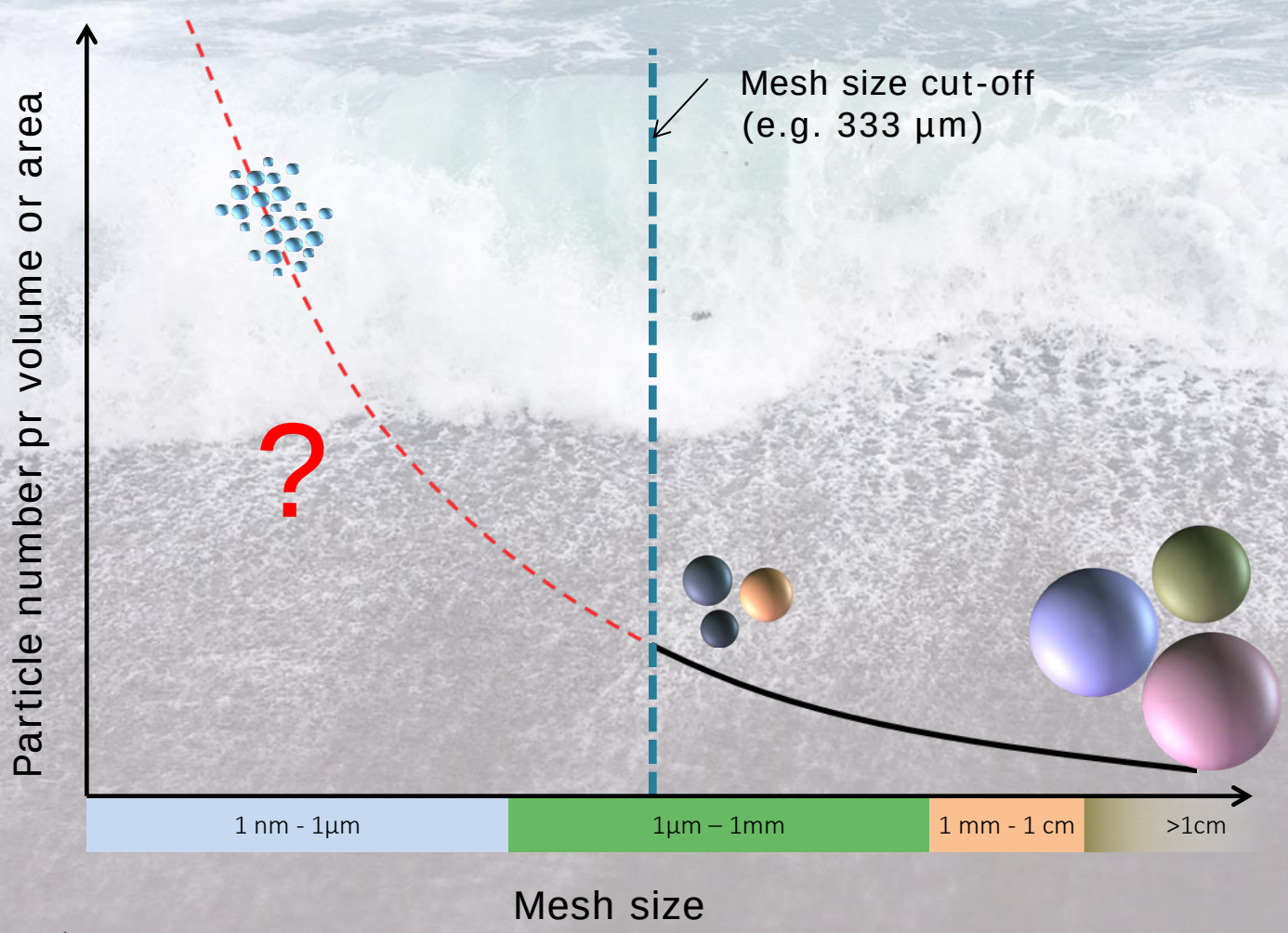


1 µm PS-COOH

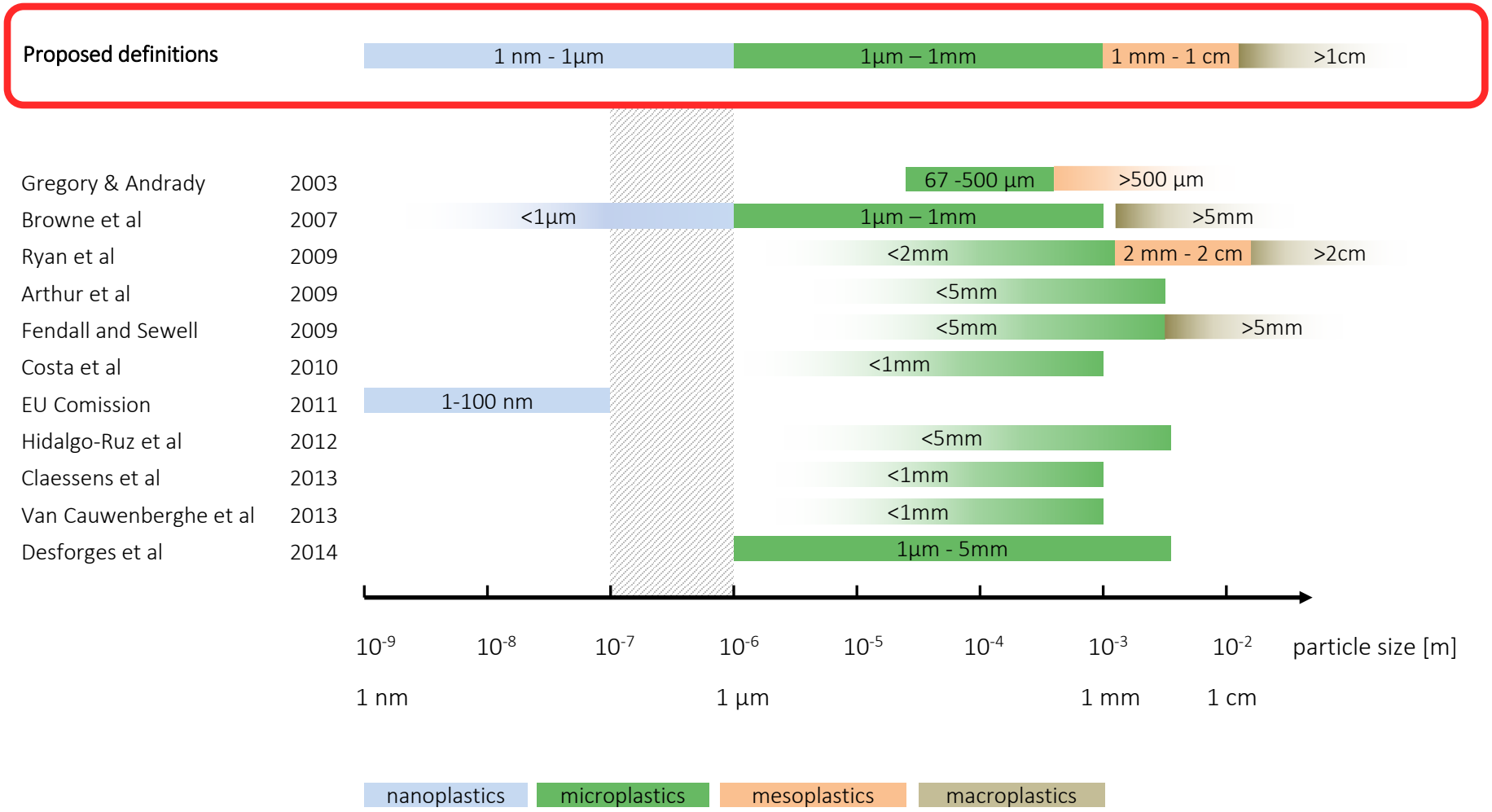


20 nm PS-COOH

Nanoplastics – an environmental problem?



Definition of nanoplastics



Some lessons learned from nanoecotoxicology…

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Interactions between nanoparticles and algae (*P. subcapitata*) by transmission electron microscopy (TEM)

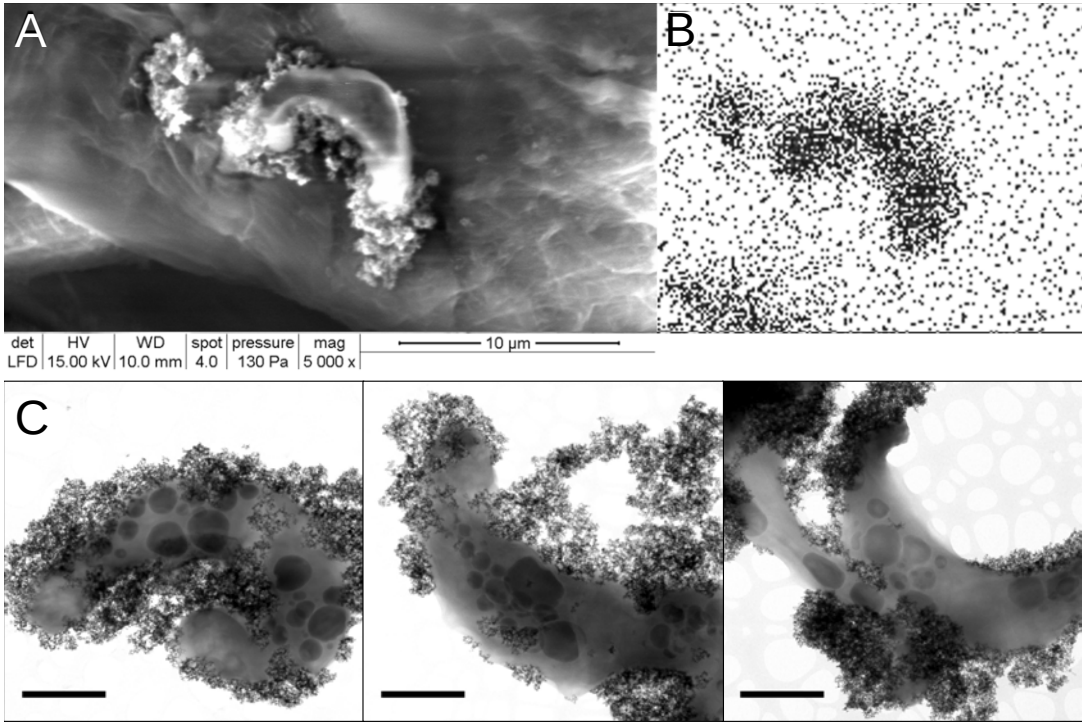
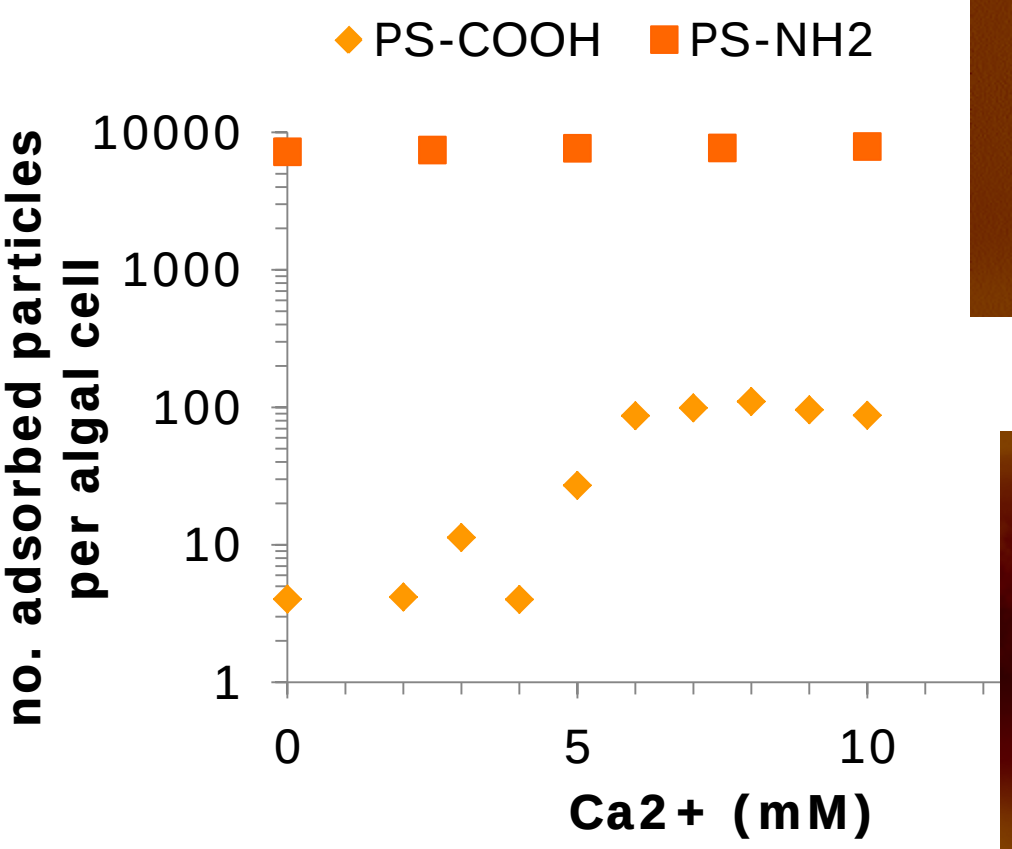
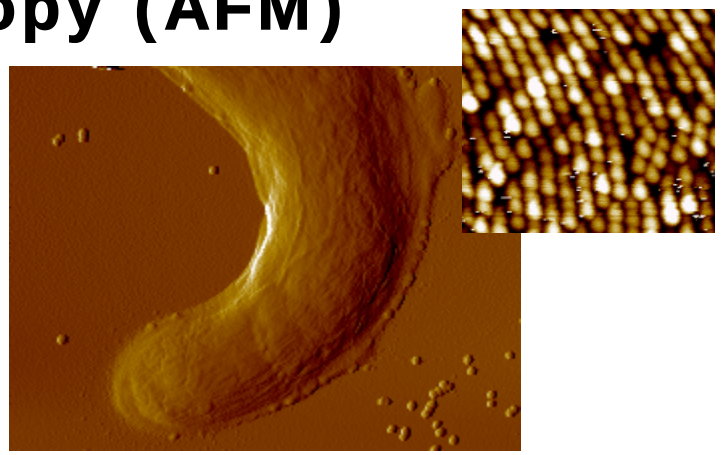
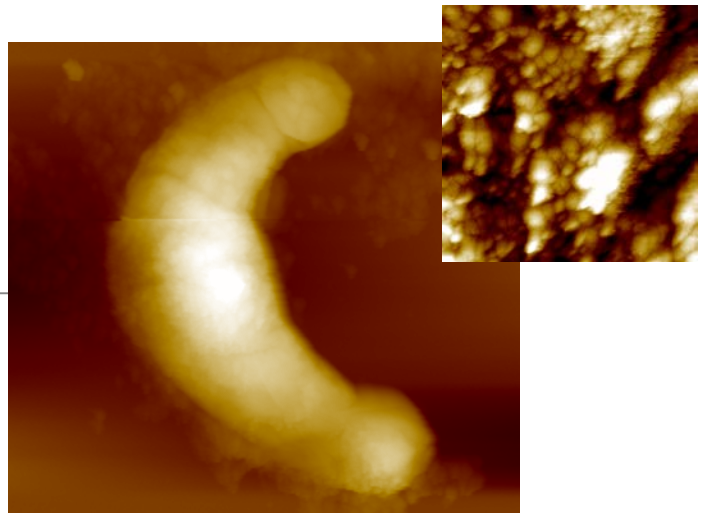


Figure 3. Encapsulation of algal cells (*Pseudokirchneriella subcapitata*) exposed to TiO_2 nanoparticles. A: Scanning electron microscopy (SEM) image of an algal cell after exposure to 50 mg/L TiO_2 in ISO algal test media for 48 h. B: Corresponding SEM-EDX dot map shows the distribution of Ti. It can be seen that the TiO_2 nanoparticles cover the surface of the algae (Modified from Hartmann et al., 2010 – Paper I). C: Transmission electron microscopy (TEM) images showing the formation of algae-particle heteroaggregates (scale bar: 2 µm). (Modified from Hartmann et al., 2011b – Paper IV)

Interactions between nanoplastics and algae cells (*P. subcapitata*) by UV-VIS spectroscopy and atomic force microscopy (AFM)







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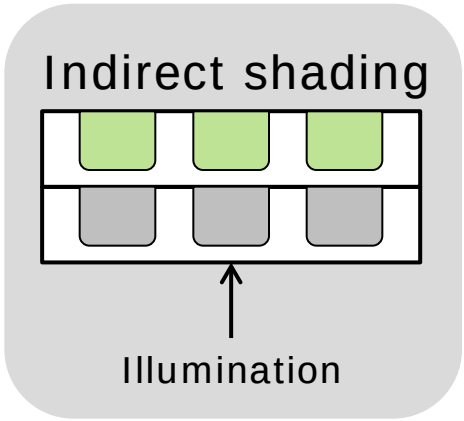
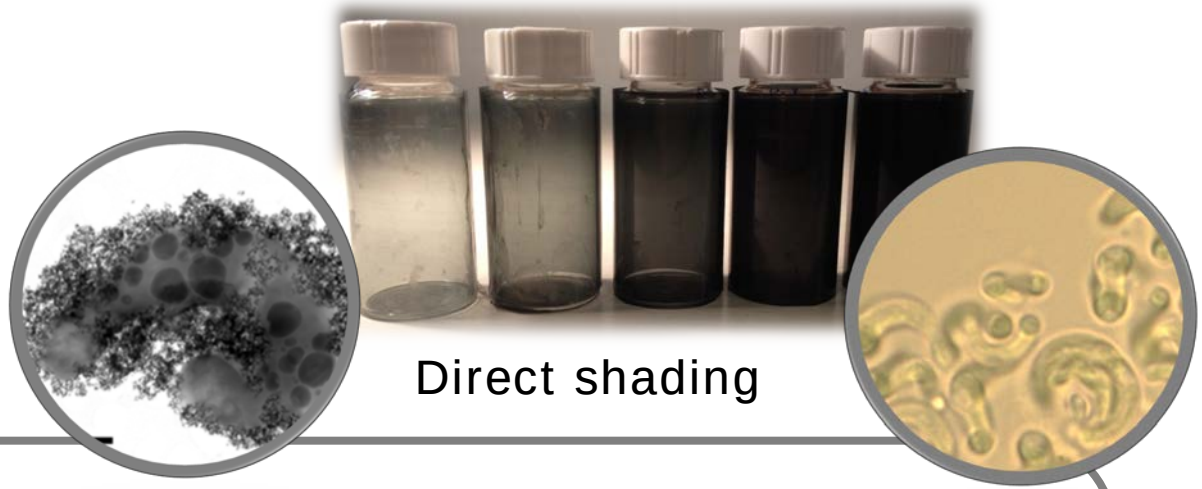
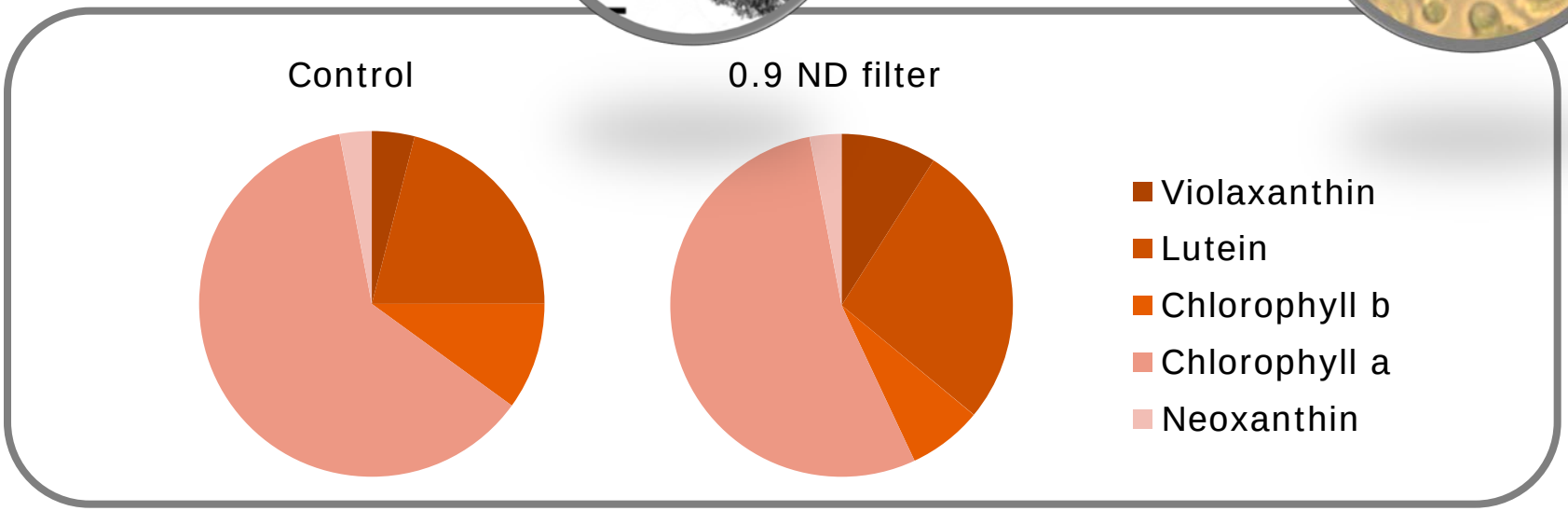
Are effects caused by shading?







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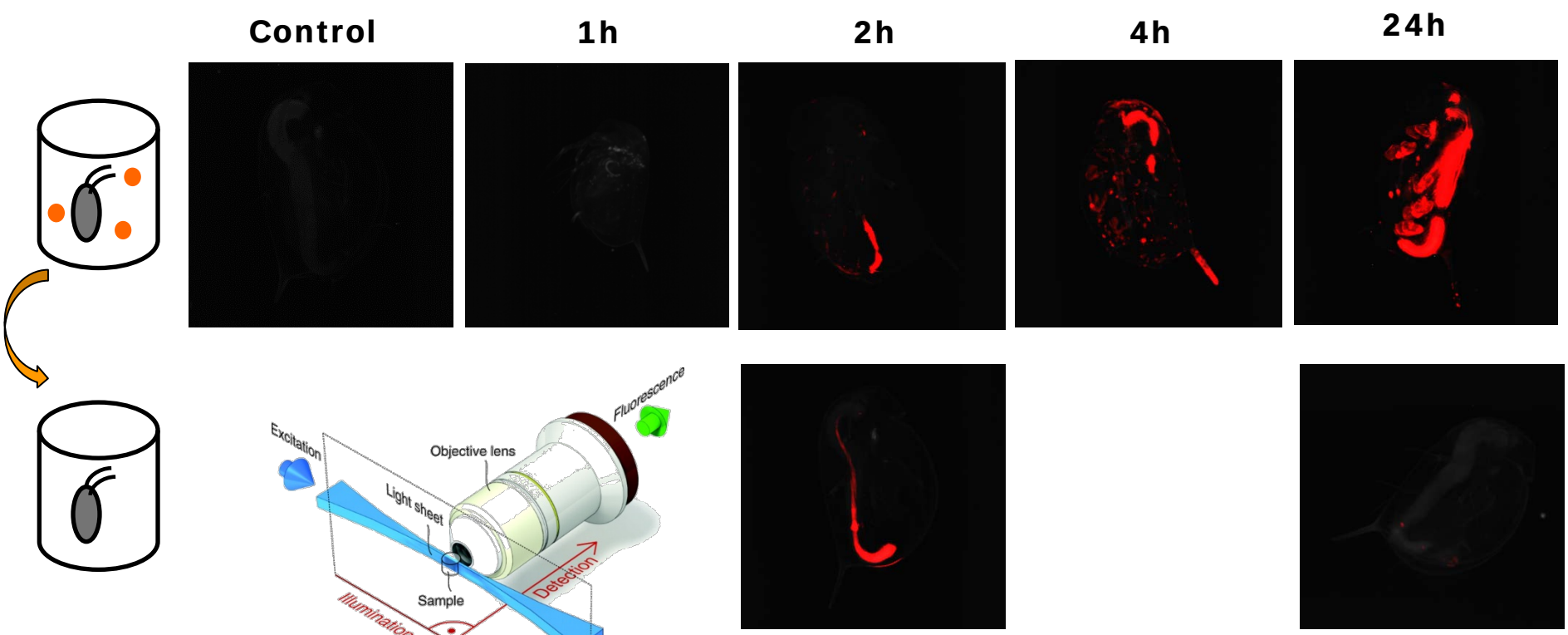
Uptake and depuration of PS beads in *D.magna*

Neonate *D. magna* (<24 h) exposed to 5 mg solids/L fluorescent polystyrene beads (20 nm). Imaged by Fluorescent Light Sheet Microscopy



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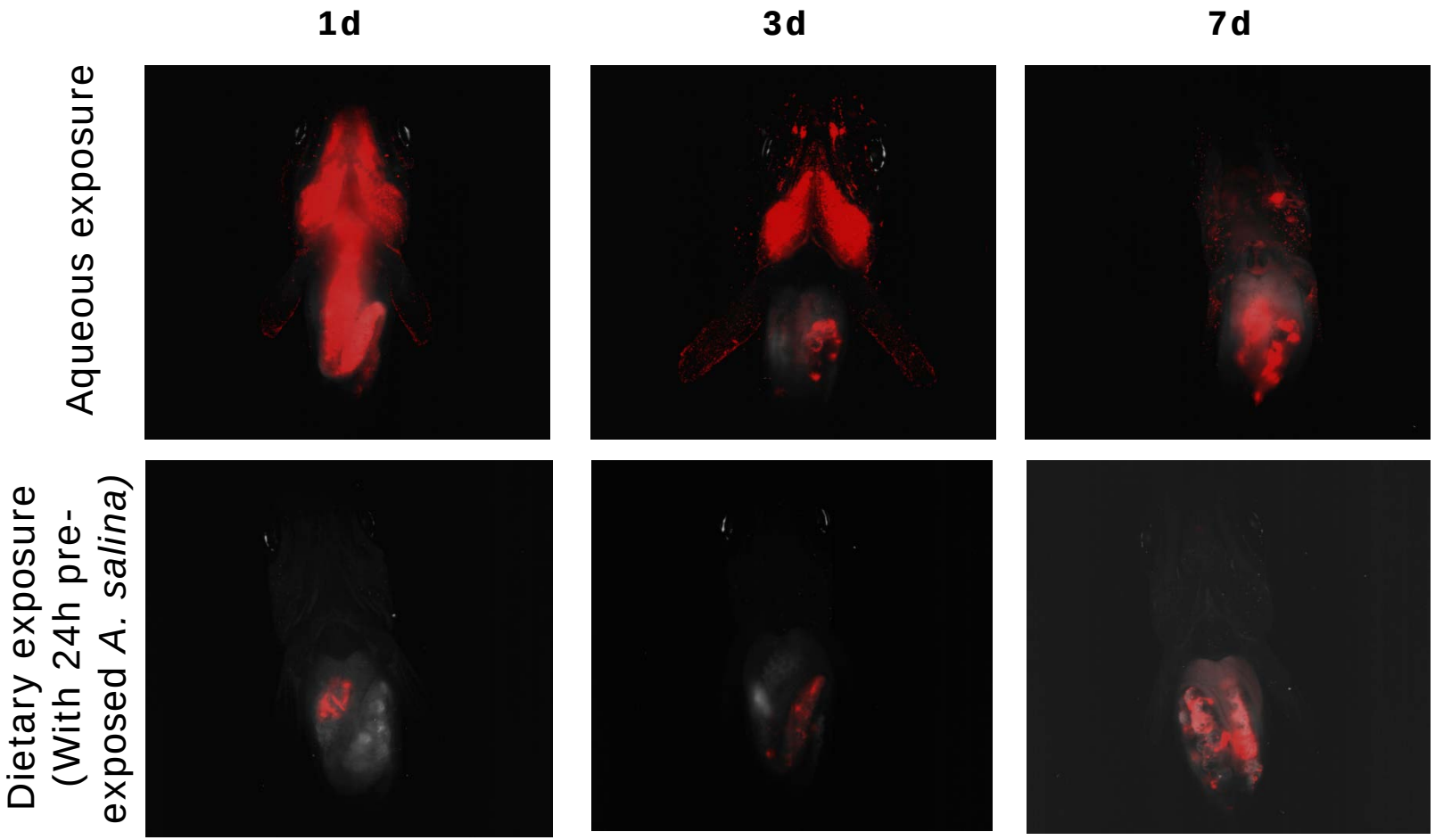
Skjolding L. M., Ašmonaitė G., Jølck R. I., Baun A. and Sturve J., 2015, Uptake and localization of fluorescent labelled nanoparticles in living crustaceans (*Daphnia magna*) and zebrafish (*Danio rerio*) using Light Sheet Microscopy, manuscript

Uptake of PS beads in juvenile *D. rerio* – aqueous and dietary exposure



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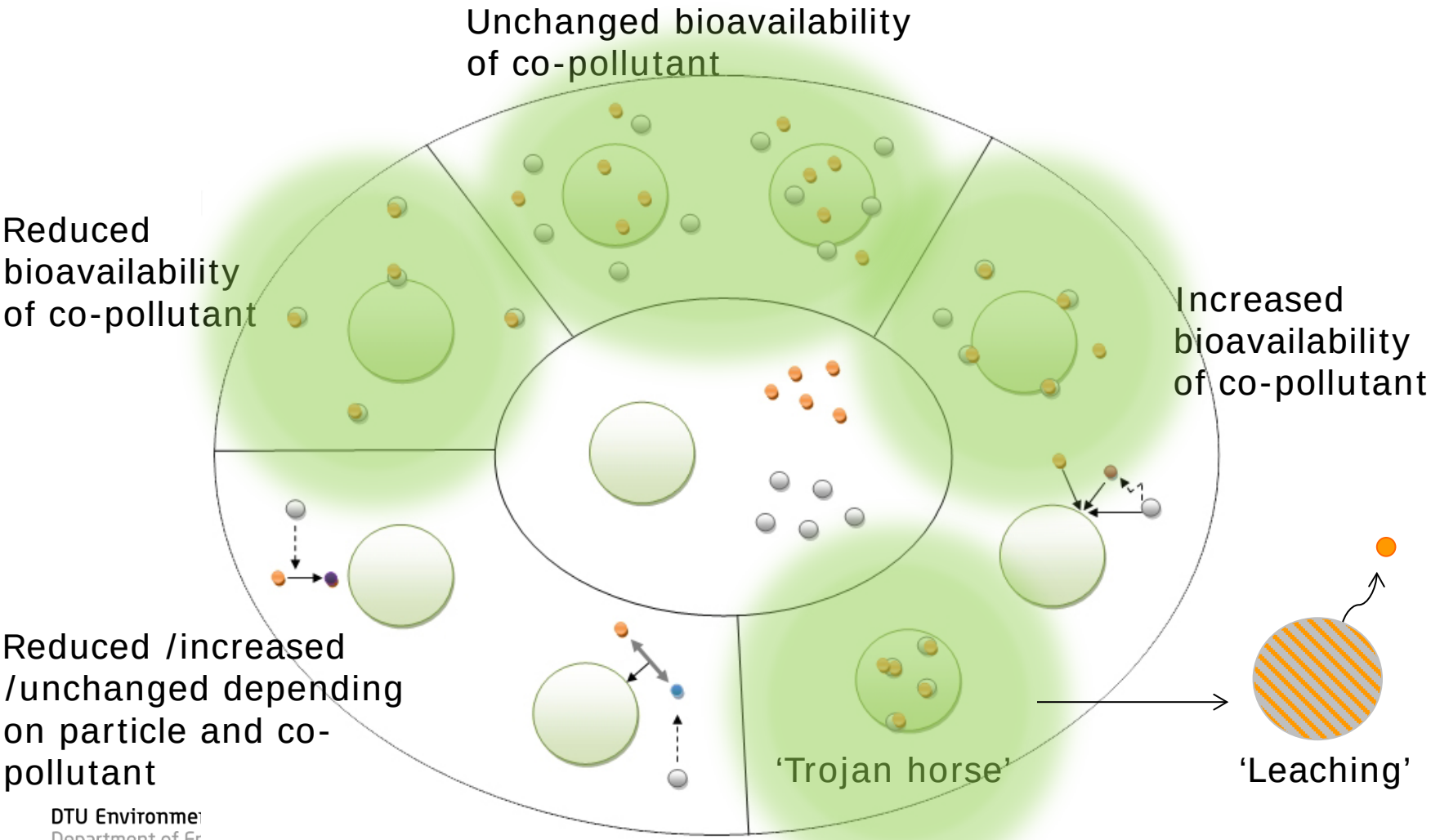
Scenarios for particle interactions with co-pollutants



Hartmann, NB, 2011, Ecotoxicity of engineered nanoparticles to freshwater organisms. PhD thesis, Technical University of Denmark, Department of Environmental Engineering, ISBN: 978-87-92654-28-1

Some lessons learned from nanoecotoxicology…

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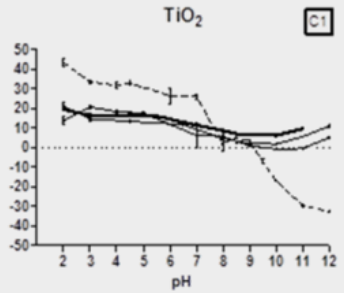
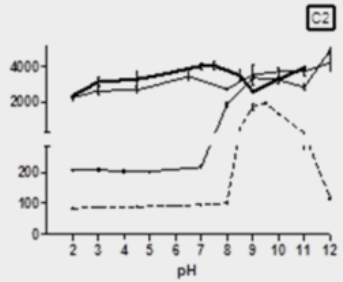
General test system considerations

- For meaningful dose-response testing we must:
  - Control exposure
  AND/OR
  - Monitor/characterise exposure

Examples:

- Dispersion method matters!
- Media composition matters!
- Time matters!

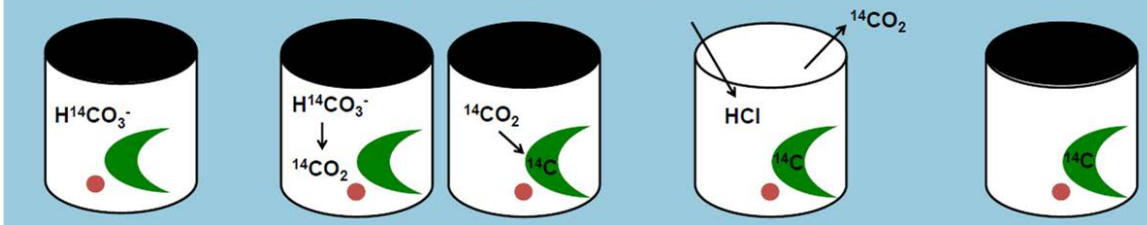




MilliQ H_2O
M7
S EPA
VS EPA

Cupi, D., Hartmann, NB, Baun, A, 2015. Influence of pH and media composition on stability and nanoparticle ecotoxicity, *manuscript*

48h test OECD 201 growth inhibition test
2h test Newly developed 2h photosynthesis inhibition test:



Test start
Algae, NPs and $H^{14}CO_3^-$ is added to media

2h exposure
Via the carbonate system $^{14}CO_2$ is formed and via photosynthesis the alga takes up ^{14}C from $^{14}CO_2$

Test end
Added HCl stops photo-synthesis and excess $^{14}CO_2$ is evaporated

Analysis
The ^{14}C amount is a measure for biomass and exposed algae is related to the control

Sørensen, S.N. & Baun, A. (2014). Controlling silver nanoparticle exposure in algal toxicity testing – a matter of timing. Nanotoxicology 10.3109/17435390.2014.913728

Exotoxicity testing of nanoplastics: some key challenges

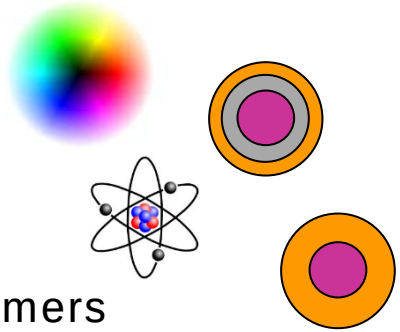
- Detection, identification and quantification in the environment
  - Properties of environmentally weathered nanoplastics?

- Quantification of exposure in laboratory test

- Detection and quantification in test organisms

Options?

- Fluorescent dye
- Metal core (synthesis)
- C14 labelled?
- Chemical analysis of polymers
  - Pyrolysis GC-MS?
  - IR spectroscopy?



Thank you for your attention!

Acknowledgements:
Lars Michael Skjolding, DTU Environment
Signe Qualmann, DTU Environment
Basil Uthuppu, DTU Nanotech
Denisa Cupi, DTU Environment
Sara Nørgaard Sørensen, DTU Environment

European Research Council Starting Grant – ENV Nano

Contact:
Nanna B. Hartmann
DTU Environment, Technical University of Denmark
nibh@env.dtu.dk