Uptake of radiolabelled carbon nanotubes (^{14}C-CNT) by the blackworm (*Lumbriculus variegatus*) – comparison of the behaviour of ^{14}C-CNT in test systems after both water and sediment spiking

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Introduction

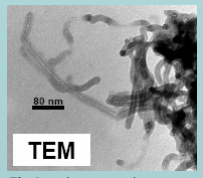
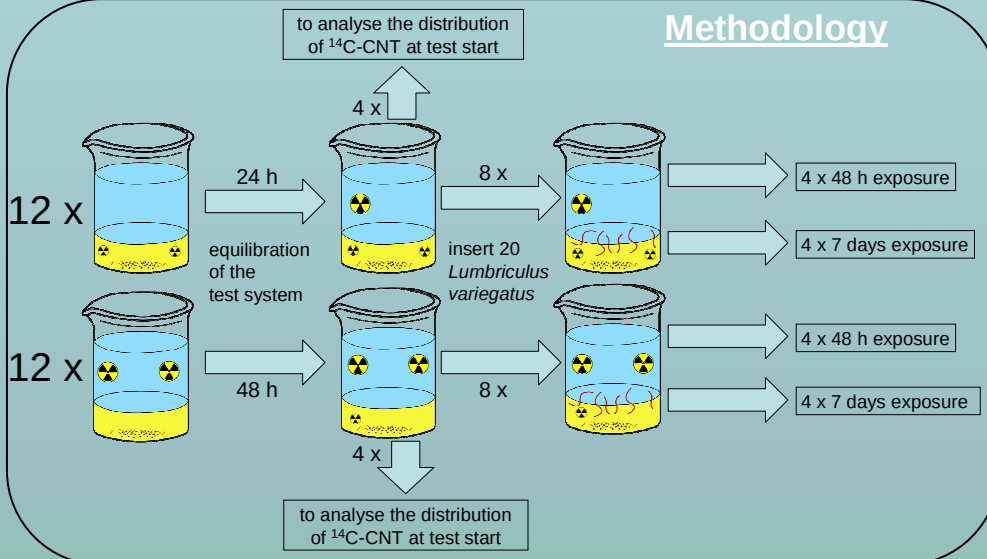


Fig.1: carbon nanotubes

Carbon nanotubes (CNT) belong to the cutting-edge materials of nanotechnology, which leads to increasing production rates. The expected widespread use requires assessment of potential risks of this material to the environment. This study deals with the bioavailability of multiwalled carbon nanotubes (MWCNT) for the blackworm *Lumbriculus variegatus* and the distribution of MWCNT in sediment-water-systems. Therefore, radiolabelled CNT (^{14}C-CNT; specific radioactivity: 1,3 MBq / mg) were used to have the possibility to quantify partitioning of CNT to the different compartments over time. Both the sediment and the water phase were spiked in order to compare these two exposure routes.


Fig.2: *L. variegatus*

Methodology



Testpreparation

sediment according to OECD TG 225

test medium according to OECD TG 203

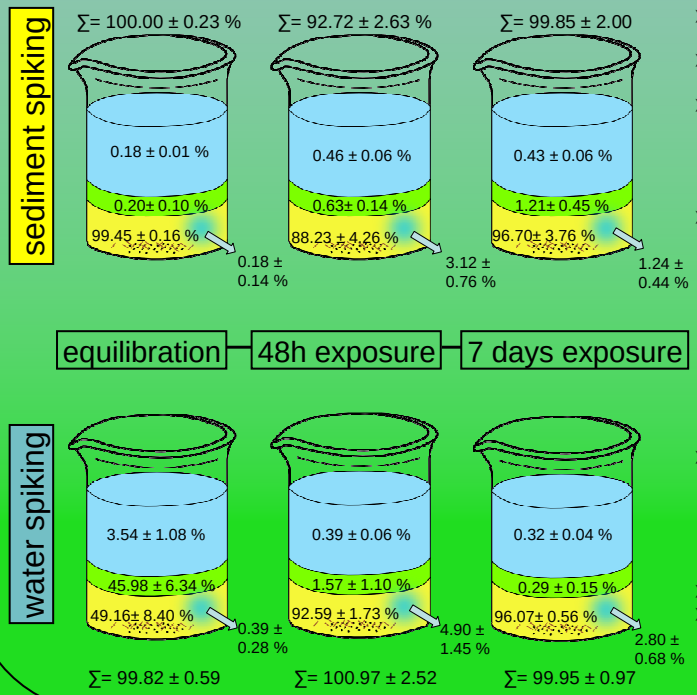
test organism: blackworm (*Lumbriculus variegatus*)

- exposure of 20 worms per test vessel (n=4)
- 10 worms sampled, dried & weighed; 10 worms purged in reconstituted water for 24 h
- homogenised in MeOH prior to liquid scintillation counting

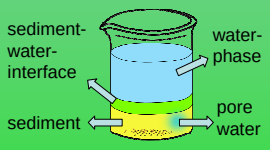
application of test material:

- 12 µg ^{14}C-CNT
- Sediment spiking: applied to 5 mL water & mixed to the other sediment constituents (pore water) → 1 µg CNT/g sediment
- Water spiking: applied to 60 mL test medium → 0.2 µg CNT/mL overlying water
- Ultrasonication with a microtip 3 x 5 min (0.2 sec pulse – 0.8 sec pause) => agglomerates and single tubes (both referred to as CNT)

Results & Discussion: distribution of ^{14}C-CNT



- little distribution from sediment to water phase
- little uptake by the organisms (< 0.4 %)
- in systems without sediment up to 2.5% was taken up from the water (data not shown), giving much higher internal concentrations
- purging of sediment-associated CNT from the gut leads to complete elimination



- clear sedimentation of CNT over time, already 46% detected in sediment-water interface after equilibration → >90% of radioactivity detected in sediment after 2 d
- little distribution to pore water
- little uptake by the organisms (<1%), but significantly more after 7 d compared to sediment exposure

Results & Discussion: uptake and elimination

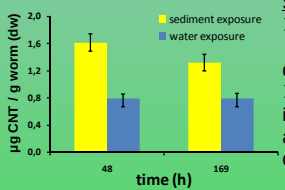


Fig.3: uptake of CNT in *L. variegatus* after 2 & 7 d exposure

Sediment spiking:
- ^{14}C-CNT concentrations of 1.3 - 1.6 µg/g worm dw
- significantly lower internal concentrations after 2 d compared to 7 d due to higher excretion than uptake

Water spiking:
- ^{14}C-CNT concentrations of about 0.8 µg ^{14}C-CNT/g blackworm (dw)

- CNT observed in gut epithelial cells by means of TEM
- Quantitatively confirmed as radioactivity was still detected in the organisms after depuration of water exposed worms (data not shown)

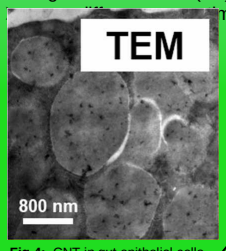


Fig.4: CNT in gut epithelial cells

Conclusions

- dispersed CNT material is bioavailable for *L. variegatus*
  - → Uptake of ^{14}C-CNT confirmed by means of TEM & ^{14}C-labelling
  - → The presence of sediment results in lower bioavailability of CNT
  - → CNT detected in gut cells if taken up from pure water - not if sediment-associated
  - → The blackworm might present a CNT-source to its predators
- MWCNT show very fast agglomeration and deposition on the sediment, which leads to low CNT exposure of pelagic organisms and potential high exposure of benthic organisms
- The use of radiolabelled CNT in aqueous dispersions leads to data sets with little scatter and enables tracking the material in all compartments of laboratory ecosystems

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