Case Study
Silver in the Environment: Mechanism of Silver Nanoparticle Toxicity

The 24th
Jyväskylä Summer School
Jyväskylän yliopisto


NANOVER
나노버 프리미엄 손청결 보습제
NANOVER PREMIUM
HAND SANITIZER
GEL
GNS
NANOVER
나노버 프리미엄 손청결 보습제
NANOVER PREMIUM
HAND SANITIZER
80ml
GNS
NANOVER
나노버 프리미엄 손청결 보습제
NANOVER PREMIUM
HAND SANITIZER
CREAM
80ml
GNS

Silver Nanoparticles (AgNPs)

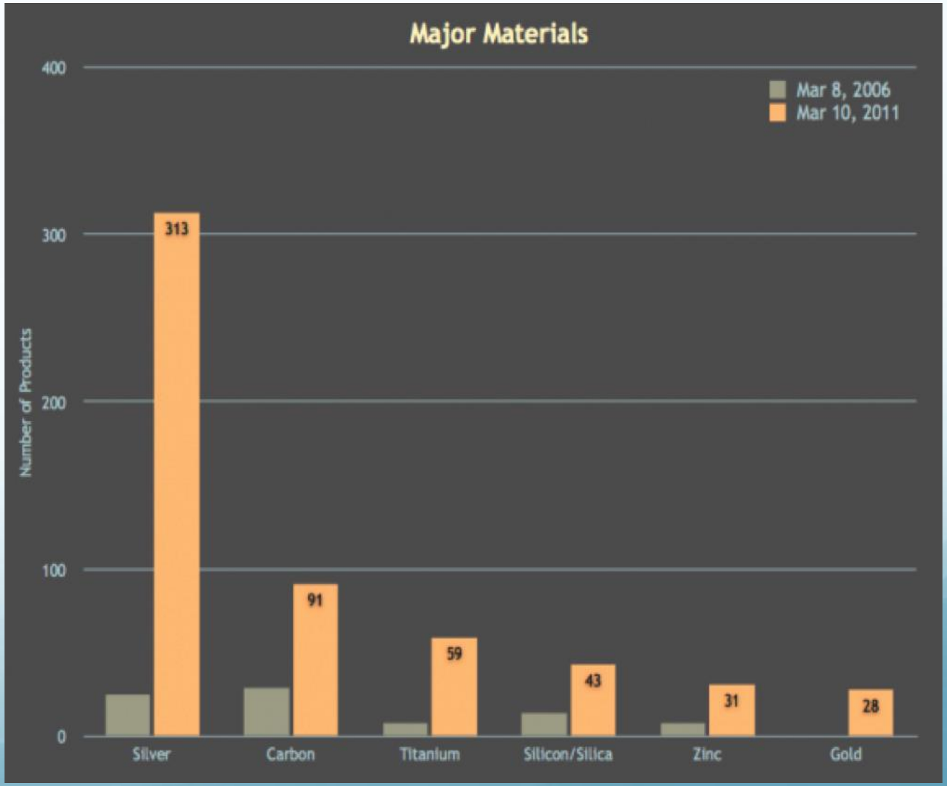
- Ag the most common material in nanotech consumer products
  - AgNPs release Ag^{+} that are antibacterial











(http://www.nanotechproject.org/inventories/consumer

Do we need to be concerned?

- Ag^{+} toxic to fishes, aquatic invertebrates

  - *D. magna* very sensitive to Ag^{+}

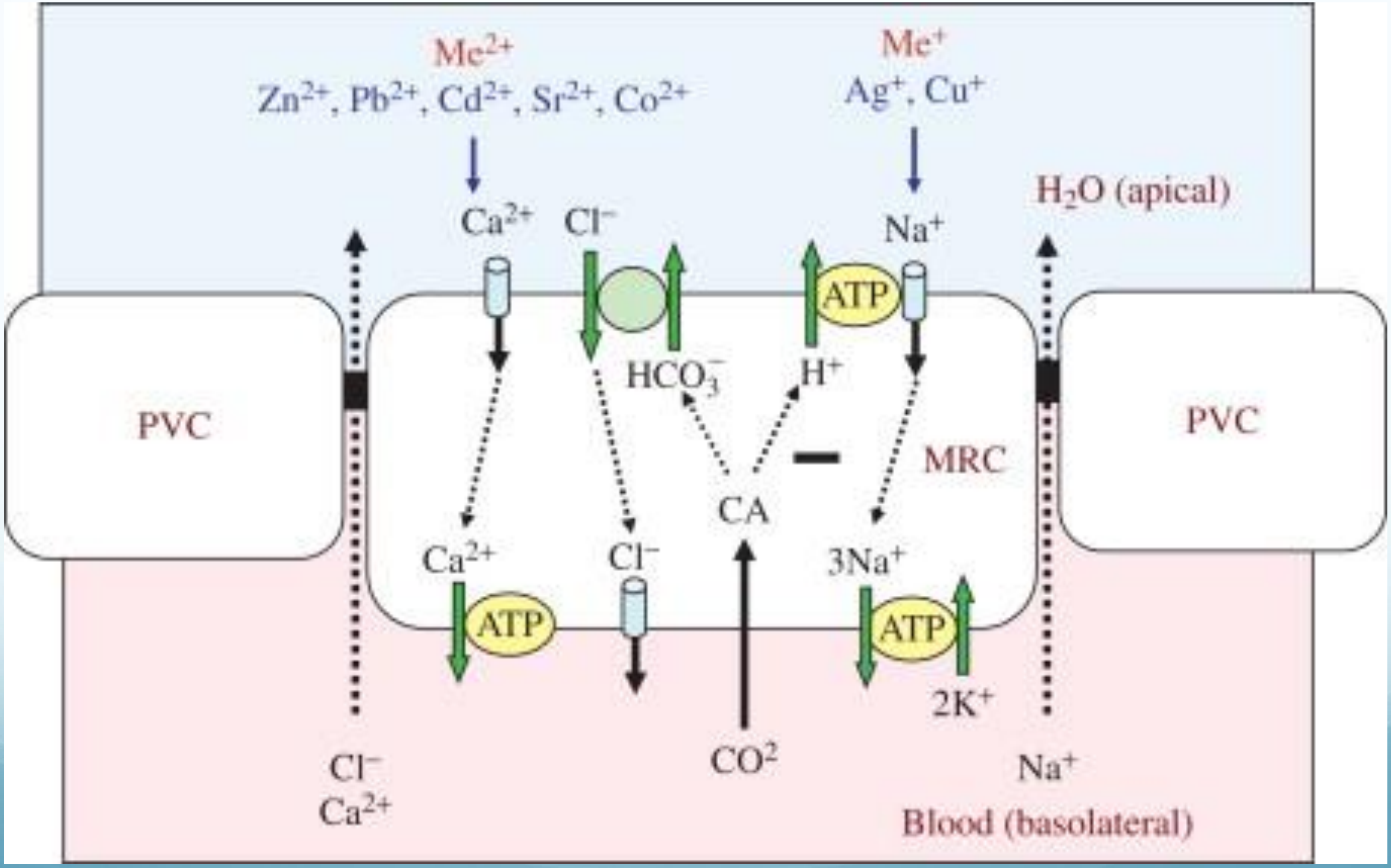
- Increased use = Potential adverse ecological impact on natural waters

  - loss of communities/populations

  - changes to food chains and ecosystem health

- Need for better understanding of the behavior of AgNPs in natural waters

Mechanism of Metal Toxicity in Fish



Wood, CM. 2011. Fish Physiology 31, Part A: 1-51

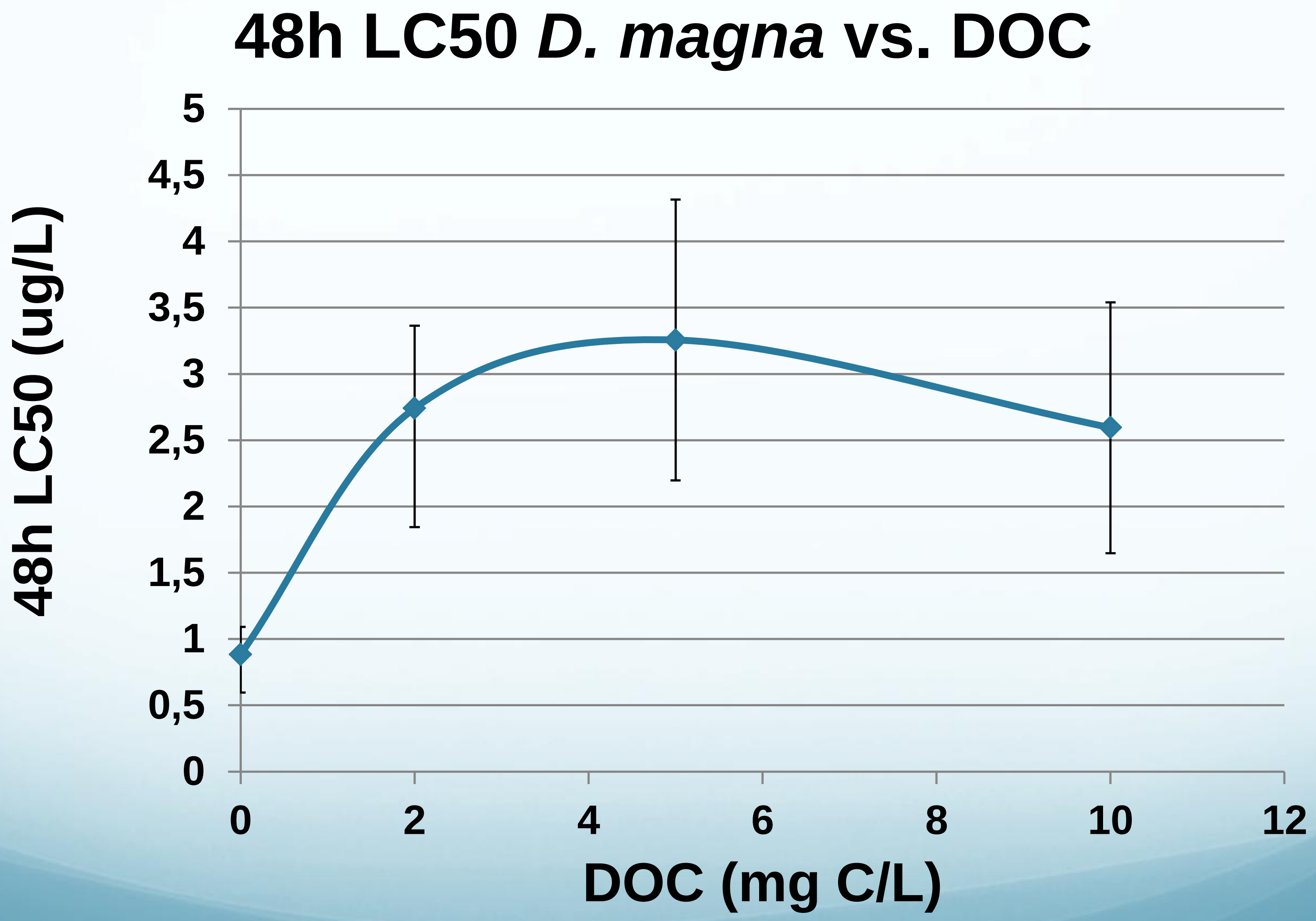
DOM is Important

- Stabilizes AgNPs: prevents agglomeration/aggregation
- Source of ligands to bind Ag^{+} and reduce bioavailability
- Studies show DOM mitigates AgNP toxicity

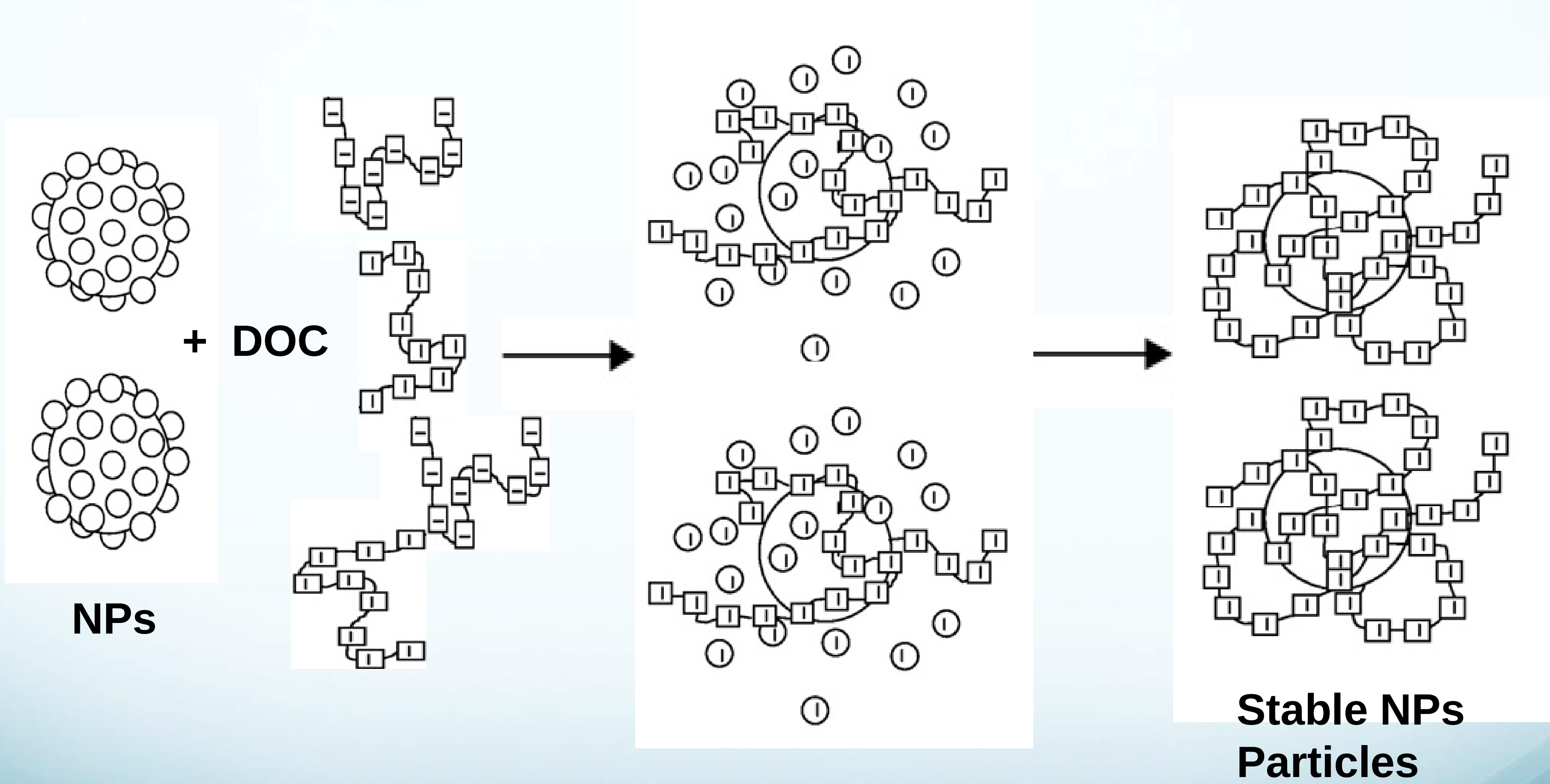
- DOC coated AgNPs released lower concentrations of Ag^{+} (Liu et al., 2010)

- DOC significantly reduced silver ion toxicity in *D. magna* (Karen et al.,1999)

Results: Karen et al.,1999



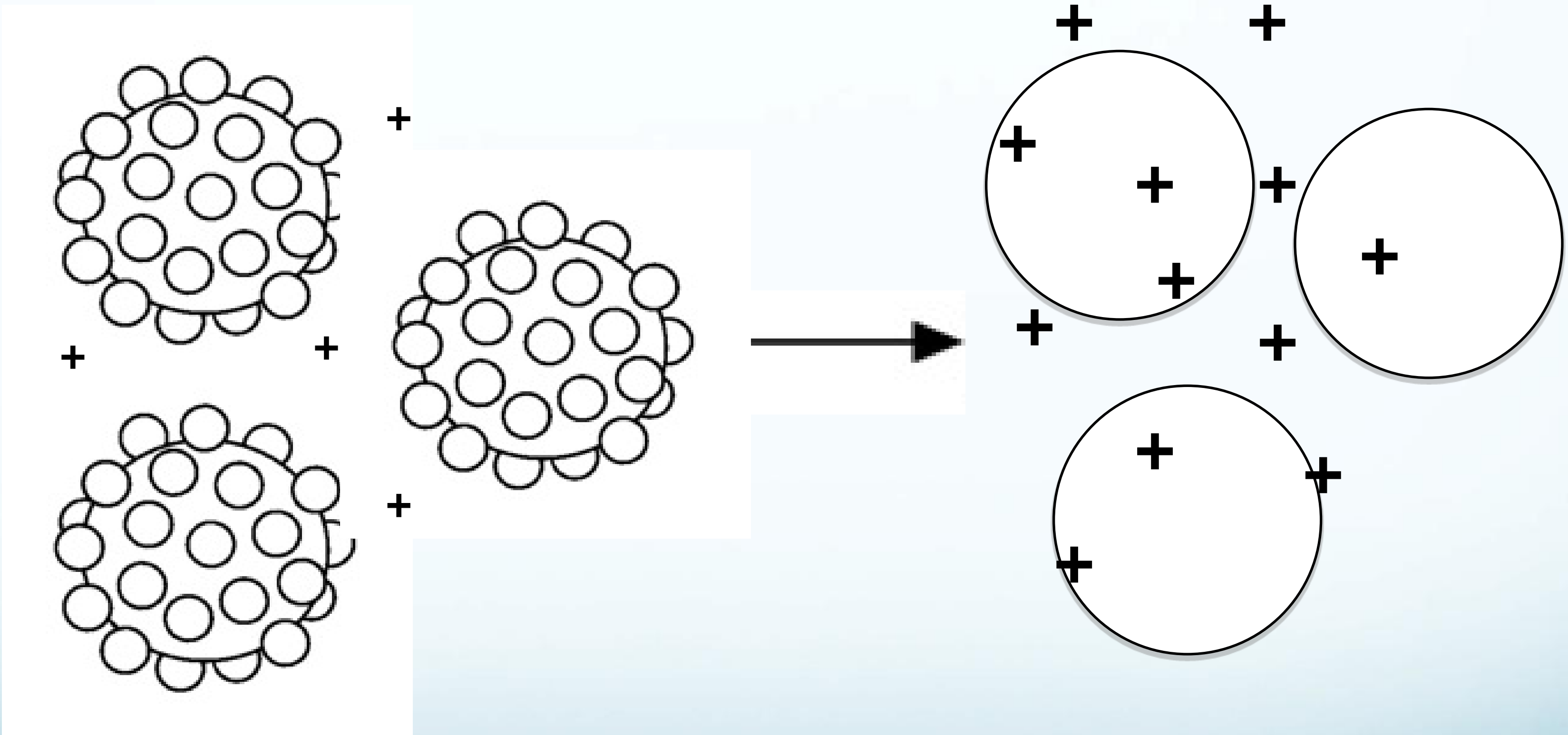
DOM Interactions



Coating is Important

- Are introduced during synthesis of AgNPs
- Are chemicals: polymers, surfactants, organic or biological molecules
- Increase stability by preventing agglomeration/aggregation and controlling dissolution
- Coatings may affect toxicity of AgNPs

Dissolution of AgNPs

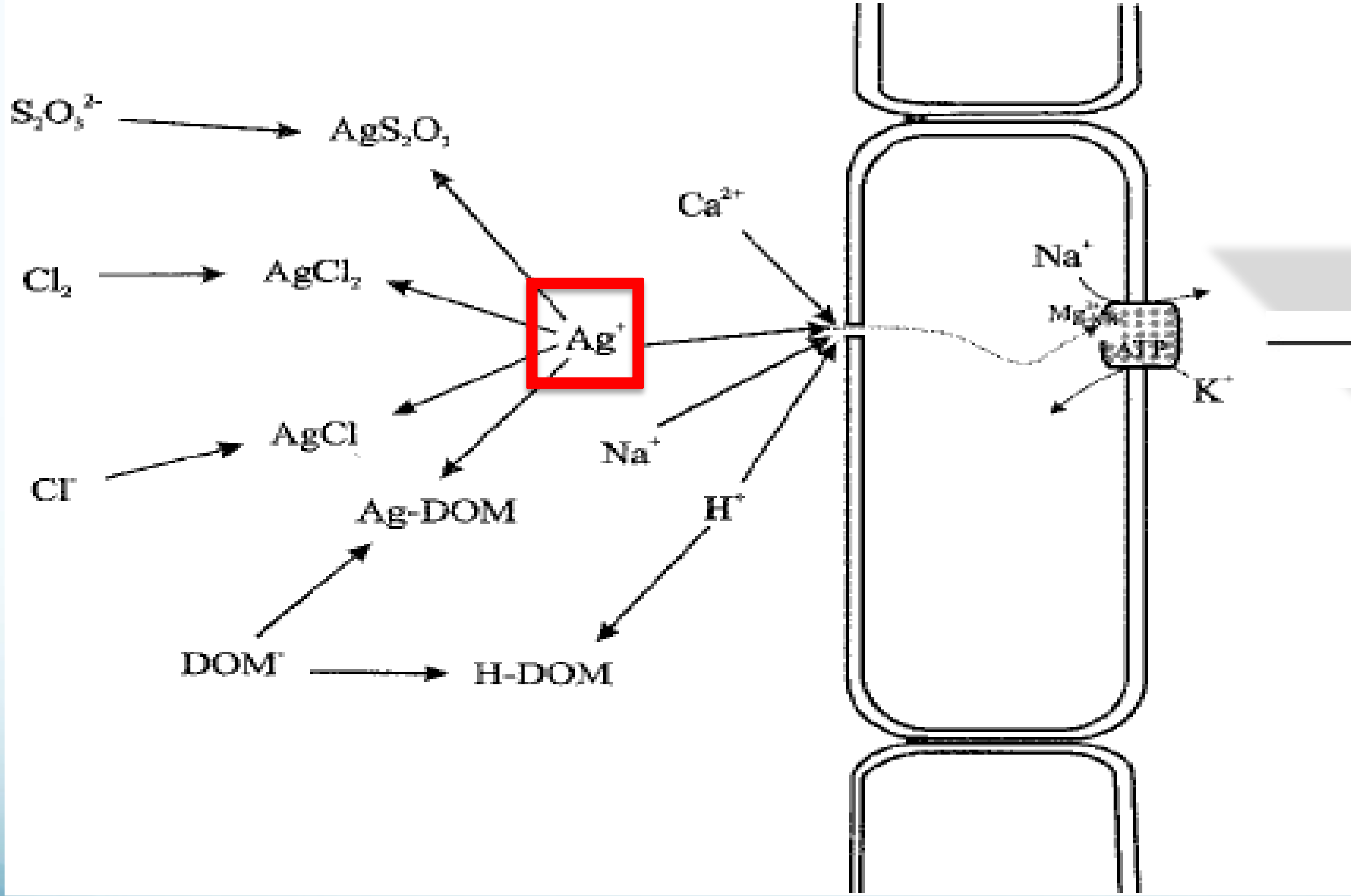


Coated AgNPs

AgNPs without Coating

Ag Speciation is important

Fresh Water Gills Blood



Complexation: abiotic ligands

Cationic competition

Biotic ligand

The questions…

- How does DOC and coatings influence toxicity of AgNPs ?

- Which is more toxic, the AgNPs, or the Ag^+ they release?

Research Goal

To characterize the toxicity of AgNPs to Daphnia magna in exposure media

Objectives

- To characterize AgNPs in exposure medium
- To conduct acute bioassays in *D. magna* with and without the addition of dissolved organic carbon in the exposure medium
- To quantify the Total and dissolved Ag at the 48h Total Ag LC50

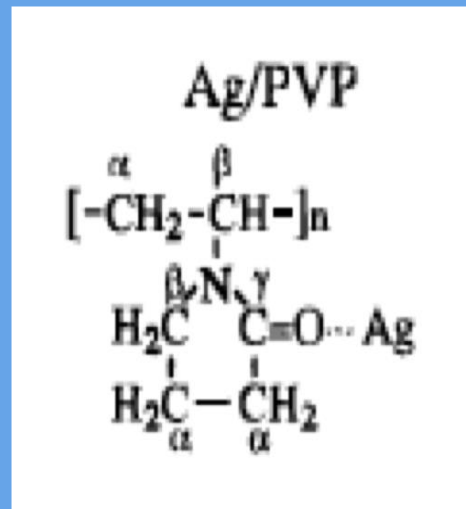
Hypothesis

The toxicity of AgNPs can be explained by the concentration of Ag^+ produced by the dissolution of the nanoparticles.

Methods: Silver Nanoparticles

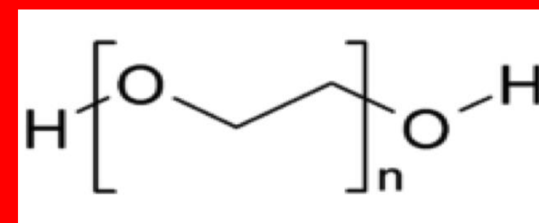
Polyvinylpyrolidone (AgPVP)

- Uncharged
- Hydrophobic region
- Hydrophilic groups
- Electrostatic adsorption to Ag
- Steric stabilization



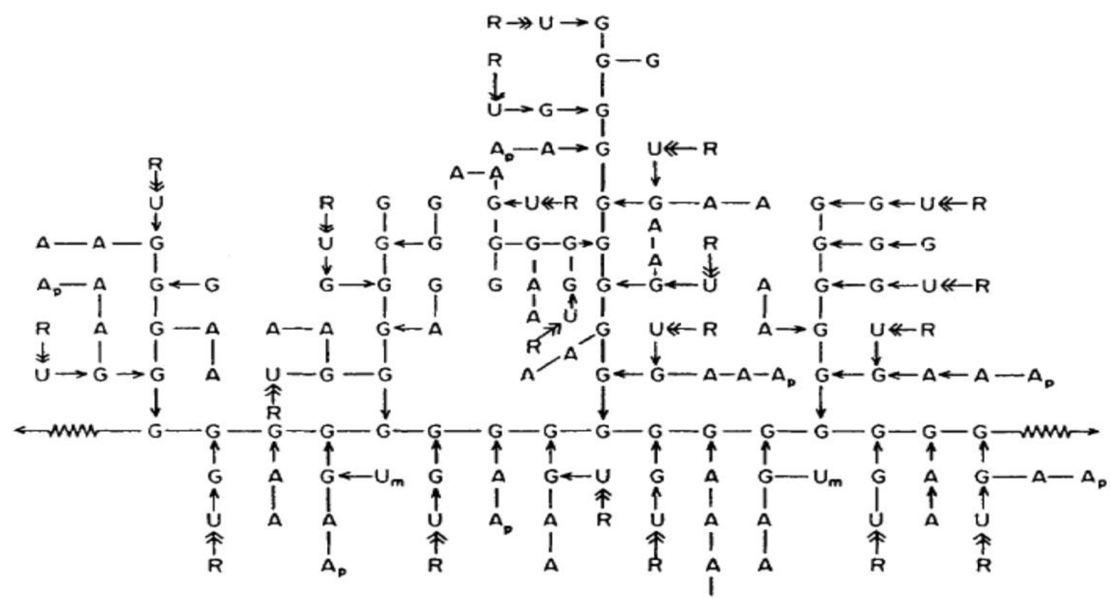
Polyethylene glycol (AgPEG)

- Uncharged
- Covalently attached to
- Steric stabilization



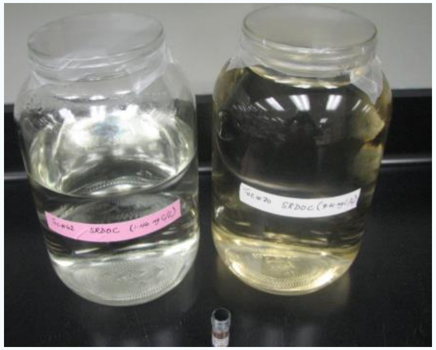
Gum Arabic (AgGA)

- Negatively charged
- Complex heterogeneous polysaccharide
- Electrostatic adsorption to Ag
- Electrostatic & mainly steric stabilization



Methods: Experimental Design

$AgNO_3$ (positive control), AgPVP, AgGA, AgPEG



0, 2, 10 mg/L Suwannee River NOM

0 + 5 – 6 concentrations

Daphnia magna

30 mL glass beakers

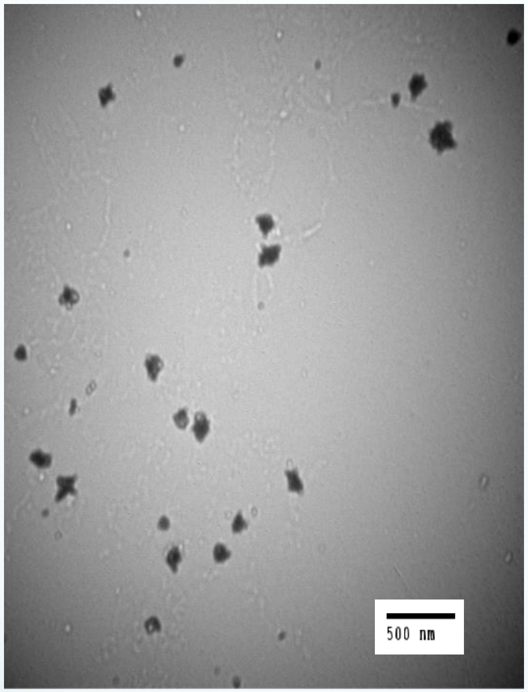
5 organisms/beaker x 3 replicates

◆ Media	EPA Moderately Hard Water
◆ Duration	48h
◆ End Point	Mortality (24h & 48h)
◆ Envirn. Conditions	25^0 C, 16h:8h Light: Dark Cycle
◆ Feeding	Fed 2h - 4h before Bioassay Not fed during Bioassay

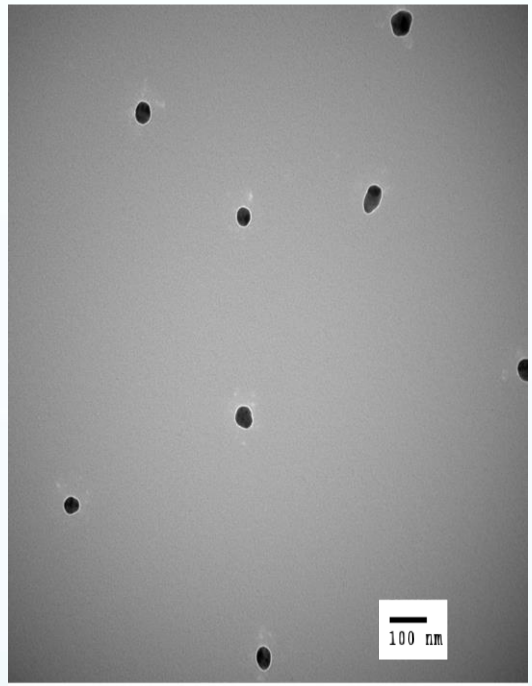
Methods: Material Characterization

- Dynamic Light Scattering
  - Hydrodynamic diameter
- Zeta Potential
  - Surface charge
- Transmission Electron Microscopy
  - Physical size and morphology

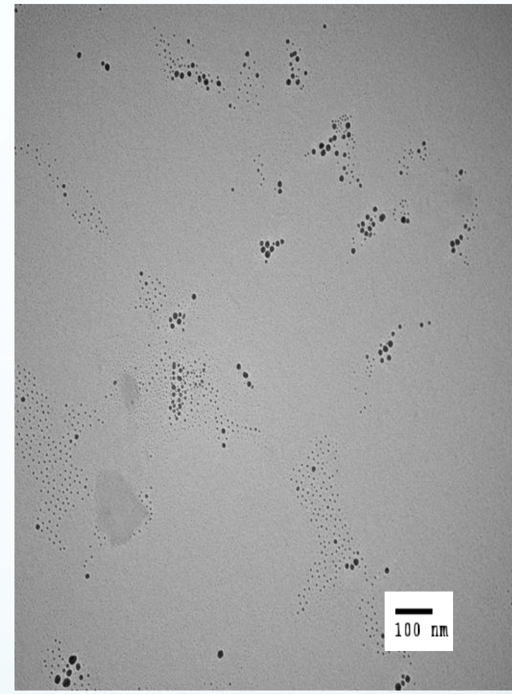
Results: TEM images of AgNPs in deionized water



(a) AgGA
Nominal: 6.0 ± 5.0 nm
Measured: 17.90 ± 7.24 nm



(b) AgPVP
Nominal: 25.0 ± 5.0 nm
Measured: 38.79 ± 9.97 nm



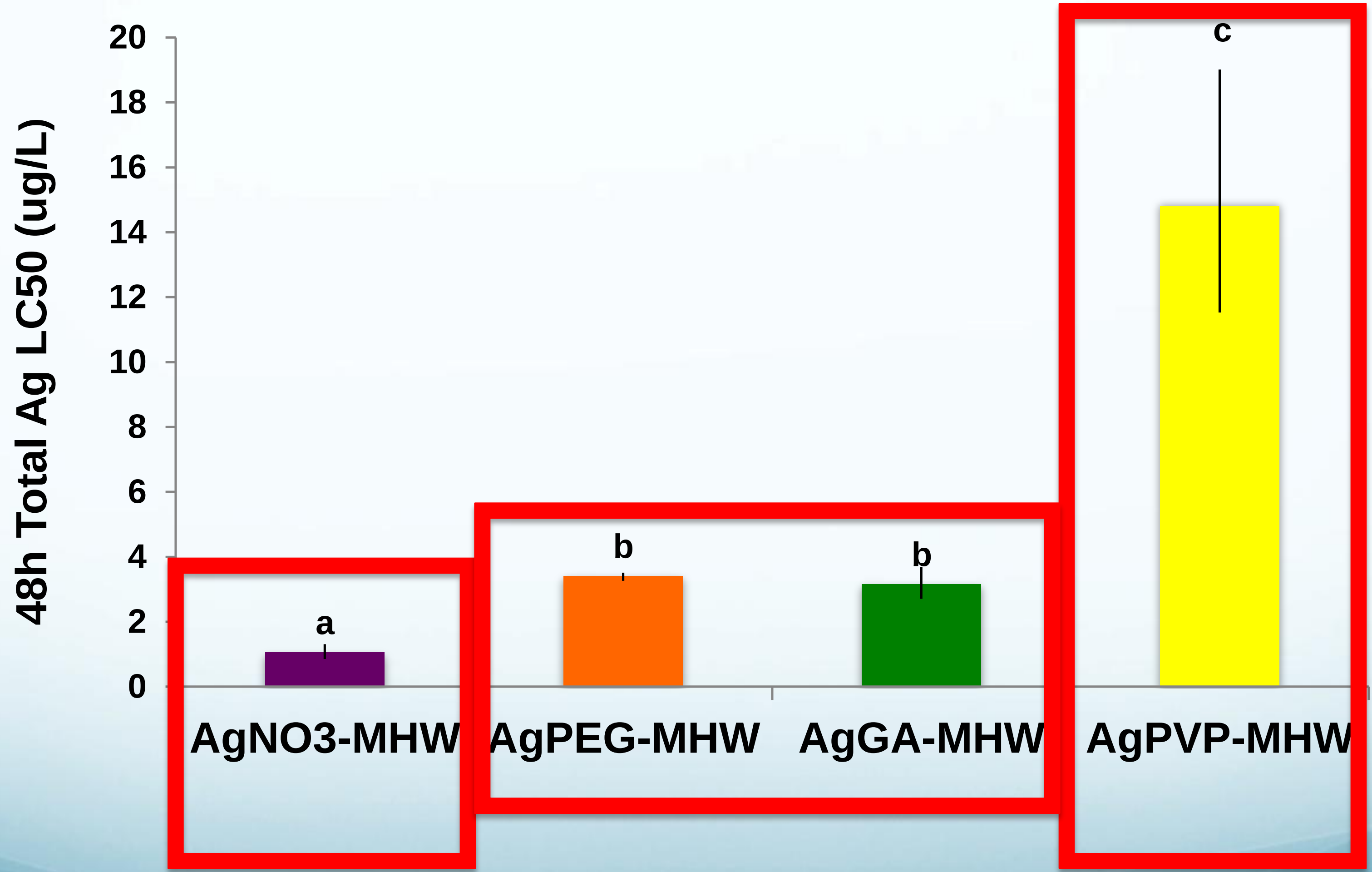
(c) AgPEG
Nominal: 4.70 ± 1.30 nm
Measured: 8.07 ± 3.84 nm

Zeta Potential -11 ± 4.11 mV

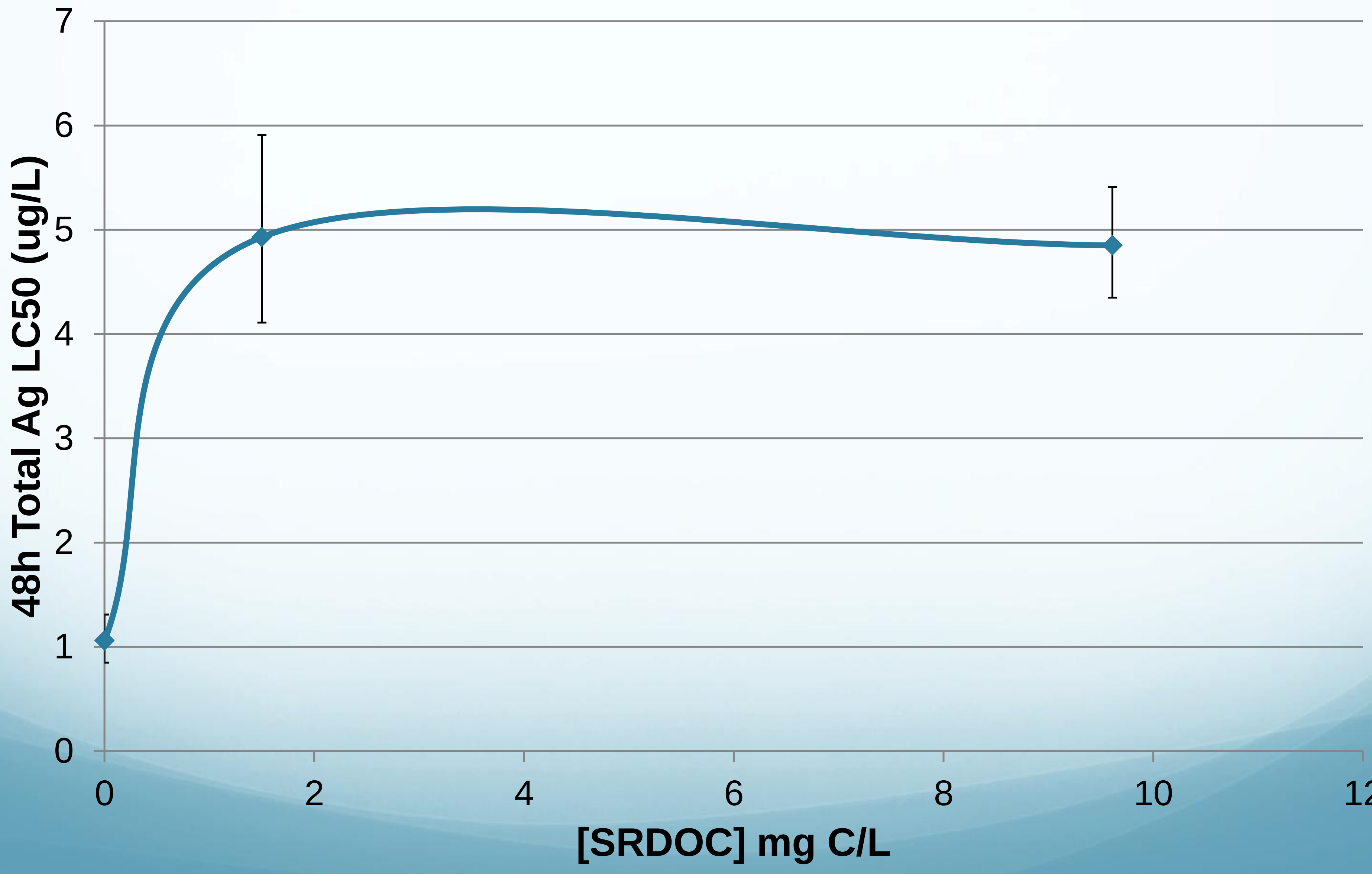
-0.103 ± 5.52 mV

-26.1± 4.39 mV

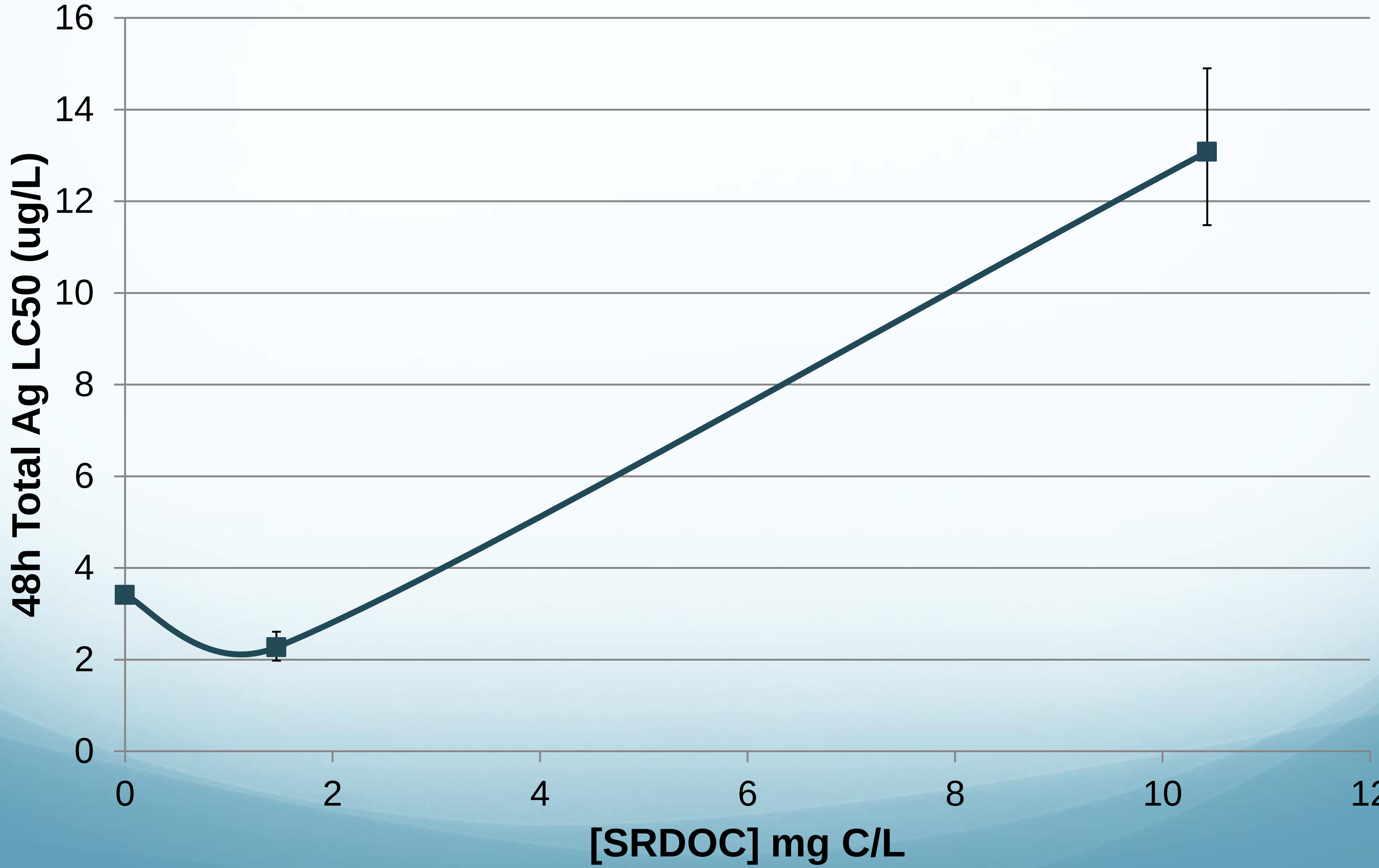
Results: Toxicity(LC50) in MHW



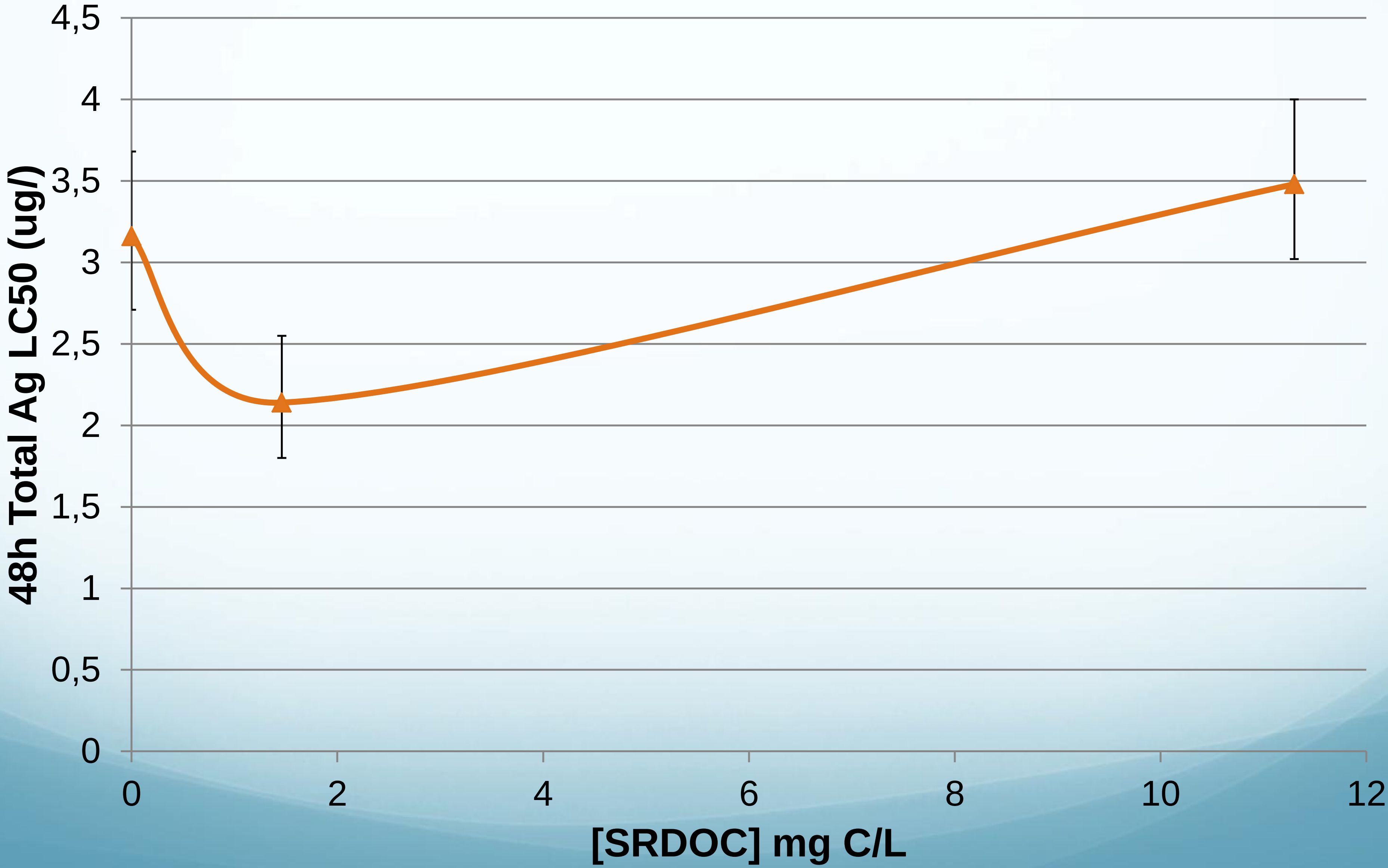
Results:Toxicity(LC50) $AgNO_3$ in SRDOC



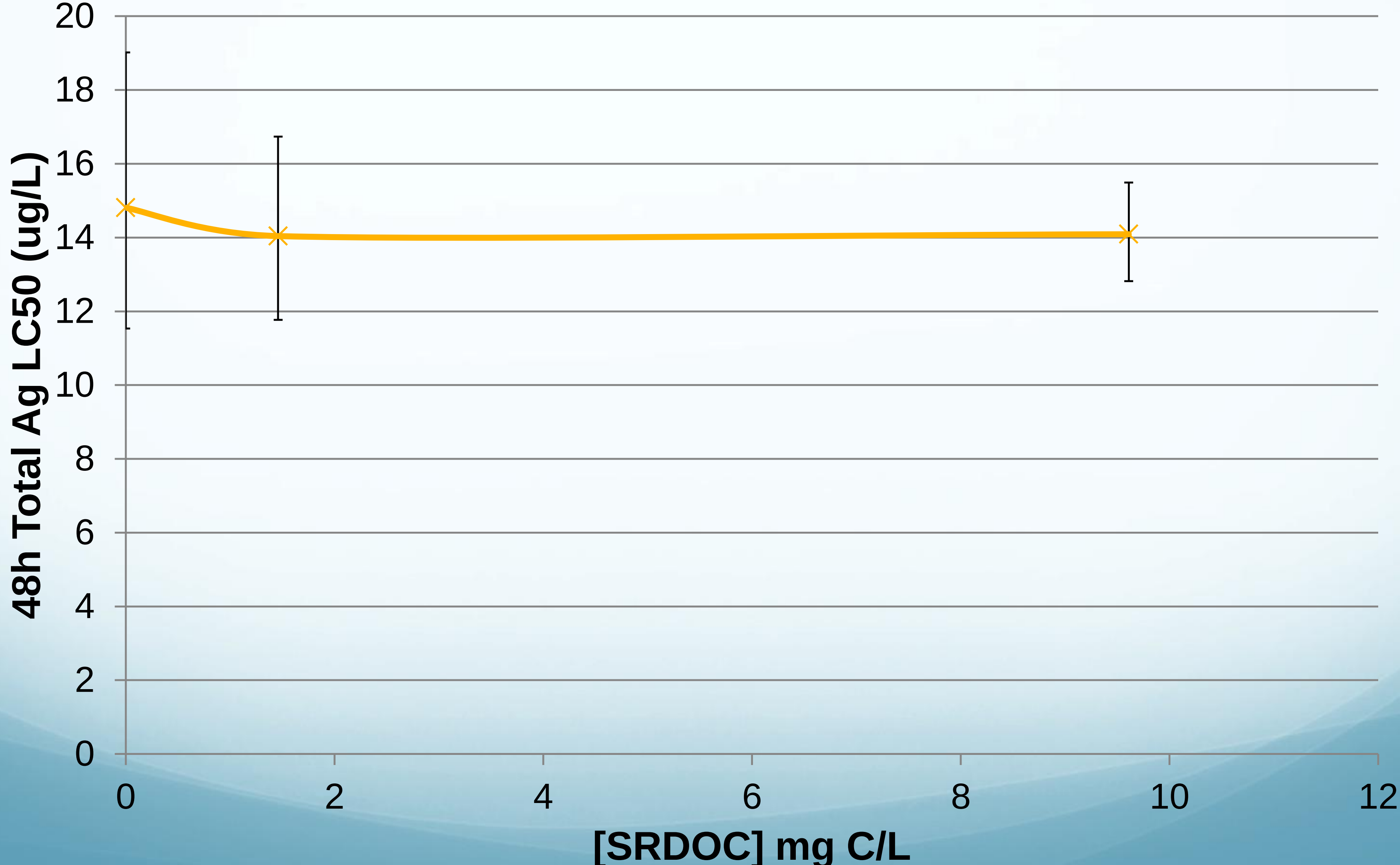
Results:Toxicity(LC50) AgPEG in SRDOC



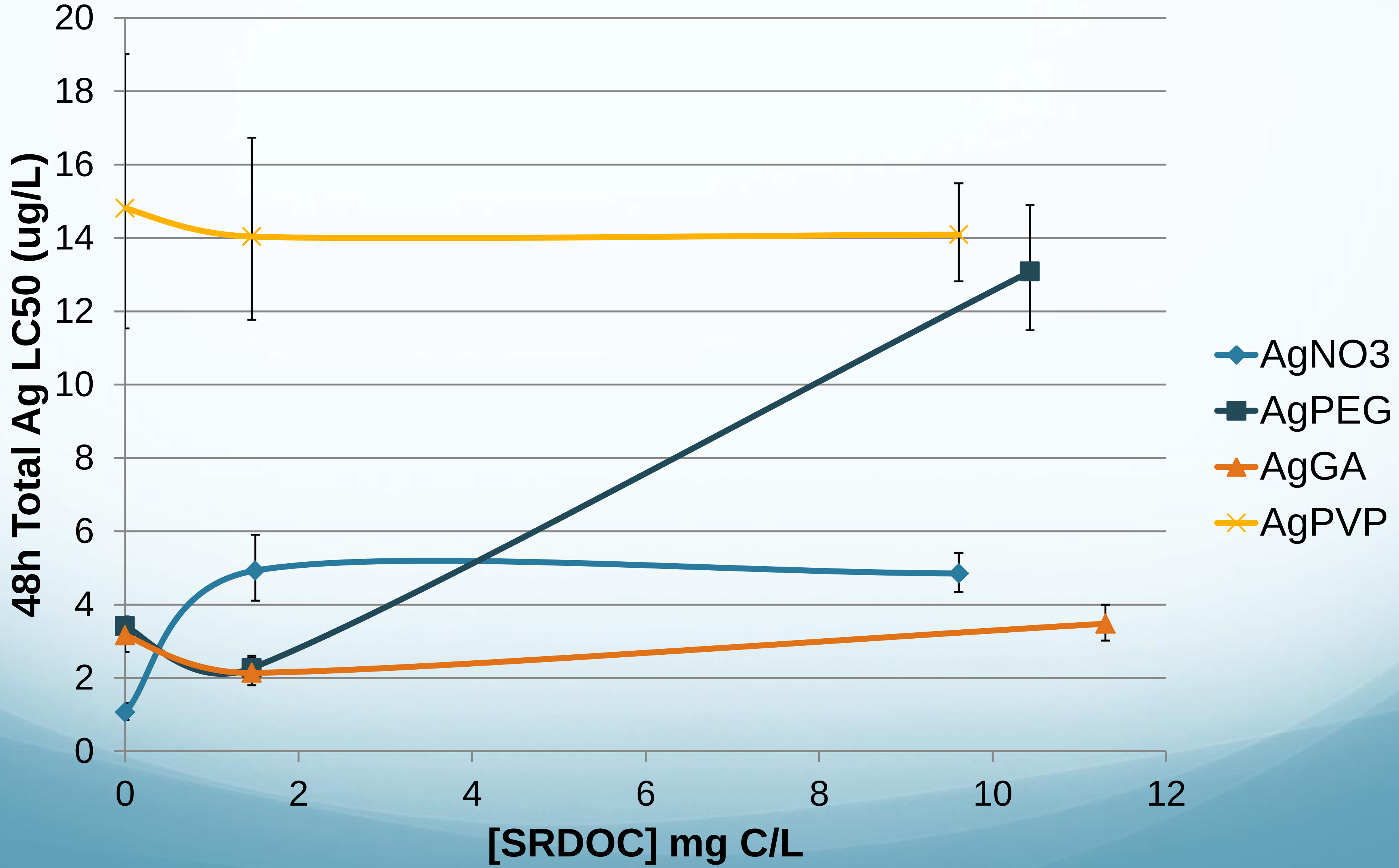
Results:Toxicity(LC50) AgGA in SRDOC



Results:Toxicity(LC50) AgPVP in SRDOC



Results:Toxicity(LC50) in SRDOC



Summary and Discussion

- In MHW AgNO3 >AgGA = AgPEG > AgPVP
- For $AgNO_3$, AgGA, and AgPEG, toxicity was significantly reduced in presence of SRDOC
- Toxicity of AgPVP was unchanged in presence of SRDOC

Objectives

- To characterize AgNPs in exposure medium
- To conduct acute bioassays in *D. magna* with and without the addition of dissolved organic carbon in the exposure medium
- To quantify the Total and dissolved Ag at the 48h Total Ag LC50

Methods: Total Ag and dissolved Ag Analysis

- Total silver analysis

  - LC50 Suspensions acidified with HCl (1%)

  - Samples analyzed using ICP-MS

- Dissolved silver analysis

  - Dissolved fraction separated by 3kDa Amicon centrifugal filter tubes

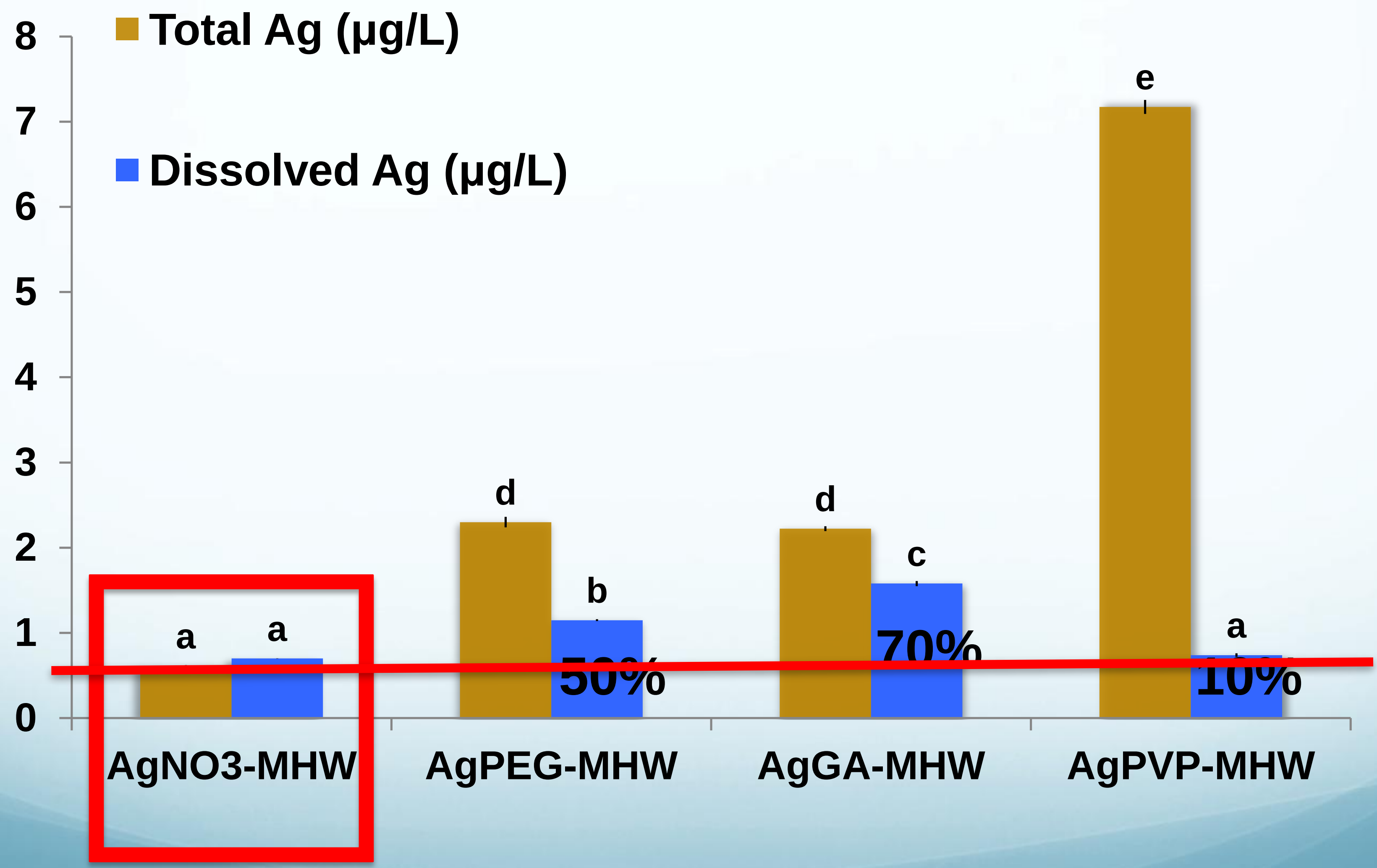
  - Residue re-suspended in fresh media then filtered

  - Final filtrate collected digested with HNO_3 (70%)

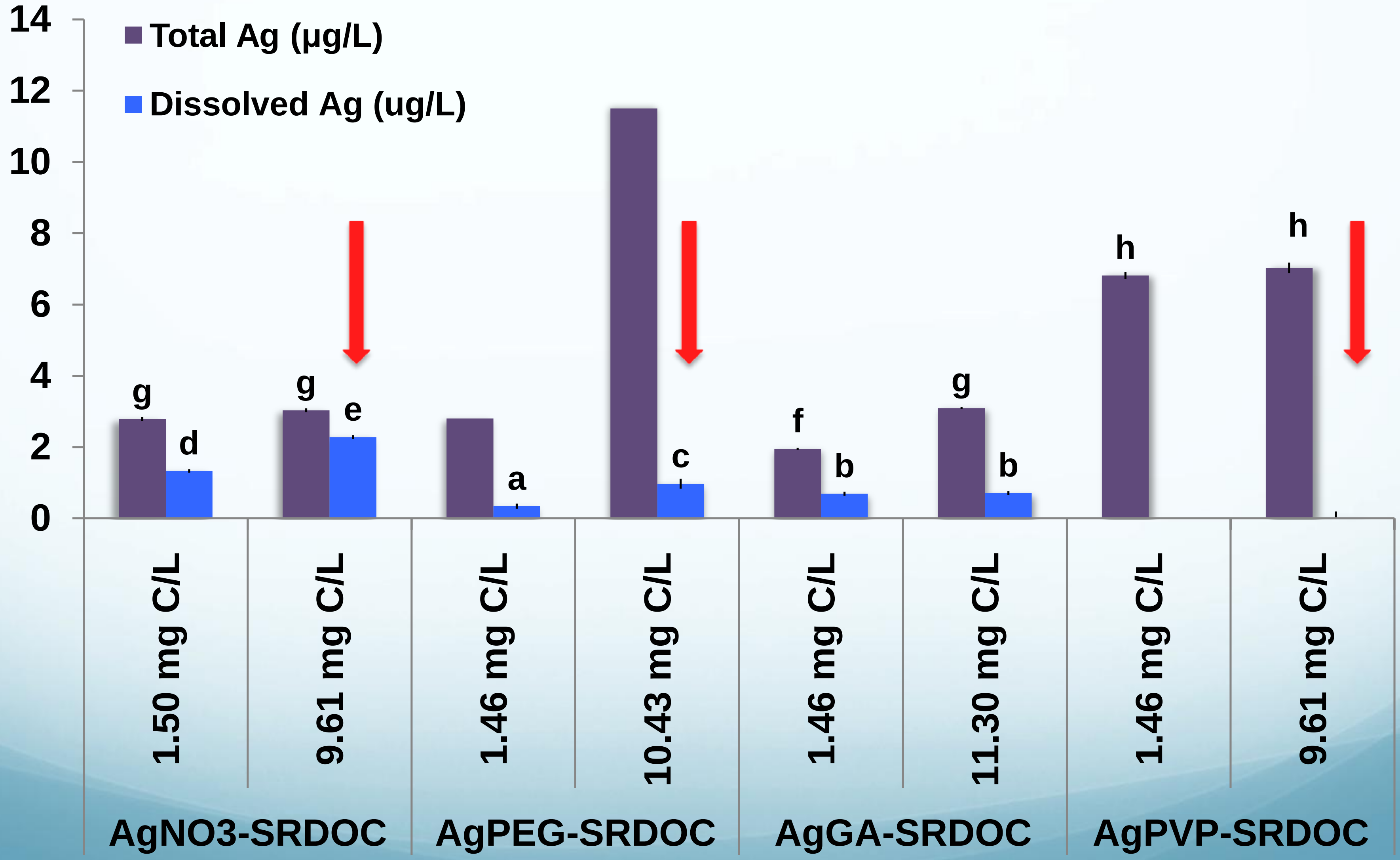
  - Diluted then acidified with HCl (1%)

  - Analyzed ICP-OES and ICP-MS

Total Silver and Dissolved Silver at LC50 for each treatment (MHW)



Total Silver and Dissolved Silver (SRDOC)



Conclusions

- The toxicity of AgNPs in MHW can be explained as a function of their respective silver ion concentrations.
- SRDOC served as a source for ligands to bind the Ag^+ and thus reduced the toxicity of the AgNPs to D. magna.