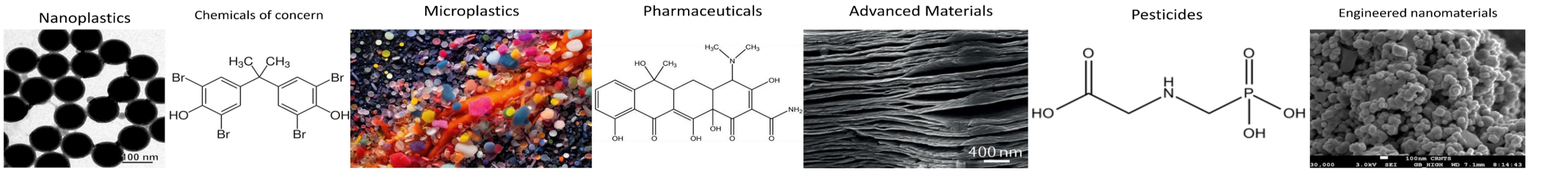
# Department Ecotoxicology
# Multiple Stressor Ecology (mStress) working group

# Effects assessment of freshwater contaminants with *D. magna*

## Background & Methods

***D. magna*** are a sensitive environmental surrogate for aquatic pollutants assessment, with protocols available for acute, chronic, behavioral and mechanistic endpoints. Also bioaccumulation and trophic transfer of pollutants via green algae can be assessed.

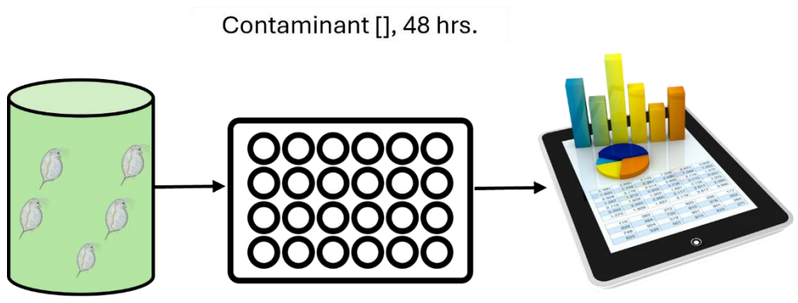



**Acute tests**
- Immobilisation (OECD 202)
- Behavioural effects
- Biochemical effects
- Internalisation

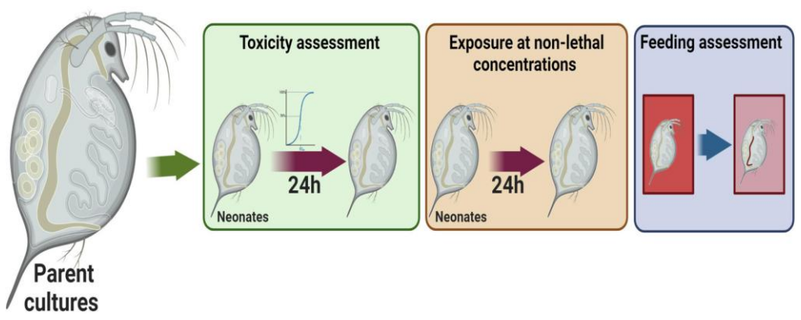
**Sub-chronic tests**
- Pollutants trophic transfer
- bioaccumulation
- Feeding rate

**Chronic tests**
- Reproduction/mortality (OECD 211)
- Growth

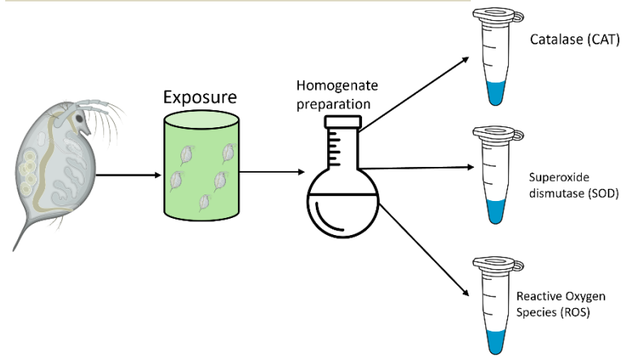
### Acute Toxicity Assay



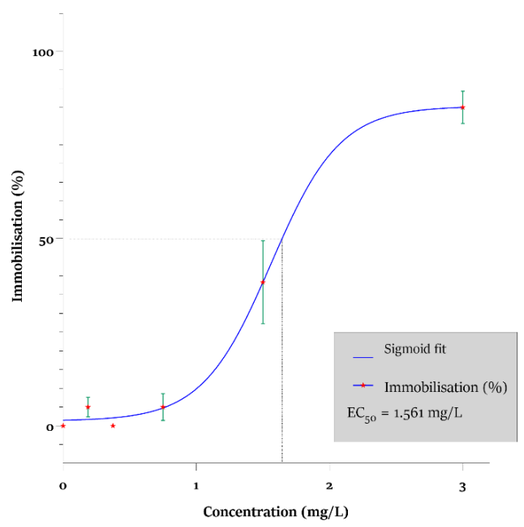

### Feeding Rate Assay



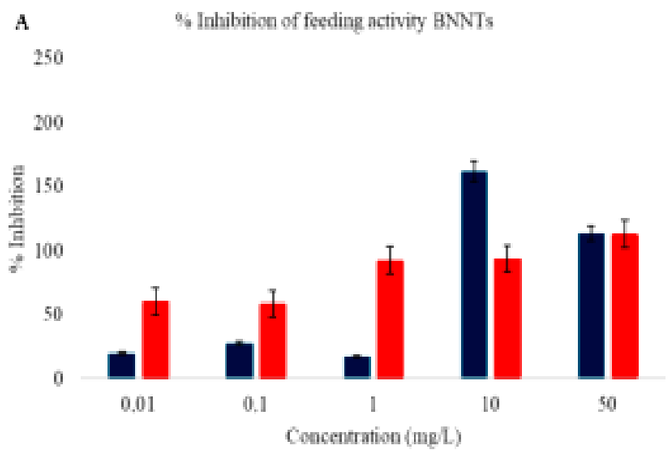

### Biomarker Assessment



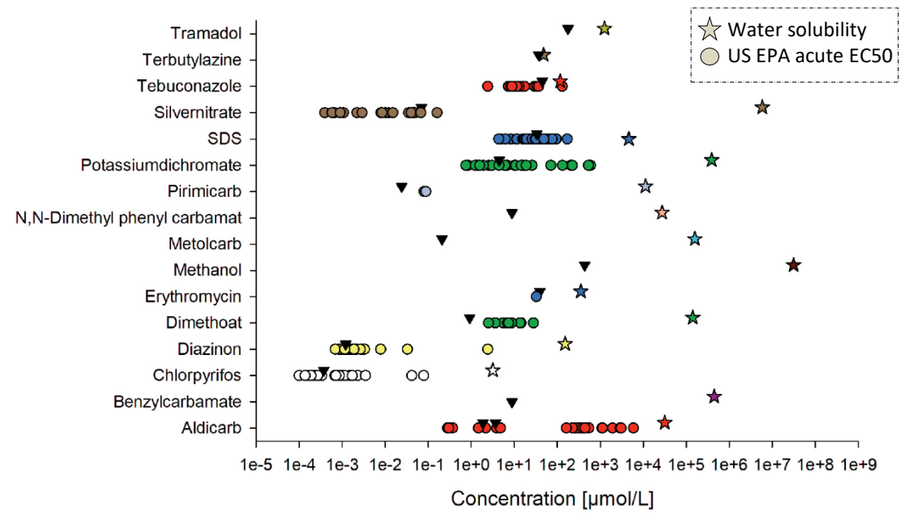

## Research Spotlights




Flame retardant TBBPA affects the **mobility** of *D. magna.*



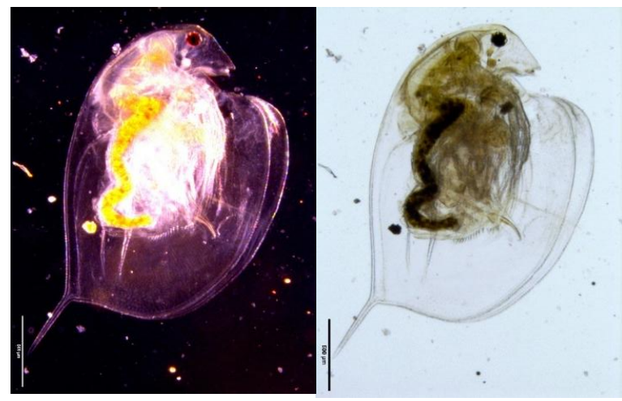

Exposure to advanced materials (Boronitrite nanotubes) affect the **feeding rate** of *D. magna.*




Acute *D. magna* immobilisation test shows comparable sensitivity in **a miniaturised approach** (▼), reducing sample volumes, increasing throughput.

## The People behind the graphs:

Dr Eberhard Küster, Senior
Dr Dana Kühnel, Senior
René Willenberg, Master student
Mbuyiselwa Moloi, PostDoc
Gail Djeukem, Master student
Silke Aulhorn, Technician



*D. magna* internalize microplastic particles, quantification of uptake?

## Key publications:


- **Moloi, M.S.**, Lehutso, R.F., Seopela, M.P., Hansen, R., Wesley-Smith, J., Motaung, L.T.T., **Kühnel, D.**, Erasmus, M., Oberholster, P.J., Thwala, M. (2025) Commercial nano-enabled products as sources of engineered nanomaterials' (ENMs) contamination in water: Release, behaviour, and ecotoxicity effects. J. Environ. Chem. Eng. 13 (5), art. 118080 10.1016/j.jece.2025.118080
- **Küster, E., Addo, G.G., Aulhorn, S., Kühnel, D.** (2023) Miniaturisation of the Daphnia magna immobilisation assay for the reliabletesting of low volume samples. UCL Open Environment Preprint 10.14324/111.444/000219.v1
- Zidar, P., **Kühnel, D.**, Sever Škapin, A., Skalar, T., Drobne, D., Škrlep, L., Mušič, B., Jemec Kokalj, A. (2024) Comparing the effects of pristine and UV–VIS aged microplastics: Behavioural response of model terrestrial and freshwater crustaceans. Ecotox. Environ. Safe. 285 , art. 117020 10.1016/j.ecoenv.2024.117020
- **Kühnel, D., Steska, T.**, Schlich, K., Wolf, C., Wohlleben, W., Hund-Rinke, K. (2023) Polymers of low concern? Assessment of microplastic particles used in 3D printing regarding their toxicity on Raphidocelis subcapitata and Daphnia magna. Microplastics and Nanoplastics 3 , art. 29 10.1186/s43591-023-00078-y
- Jemec Kokalj, A., Heinlaan, M., Novak, S., Drobne, D., **Kühnel, D.** (2023) Defining quality criteria for nanoplastic hazard evaluation: The case of polystyrene nanoplastics and aquatic invertebrate Daphnia spp.. Nanomaterials 13 (3), art. 536 10.3390/nano13030536



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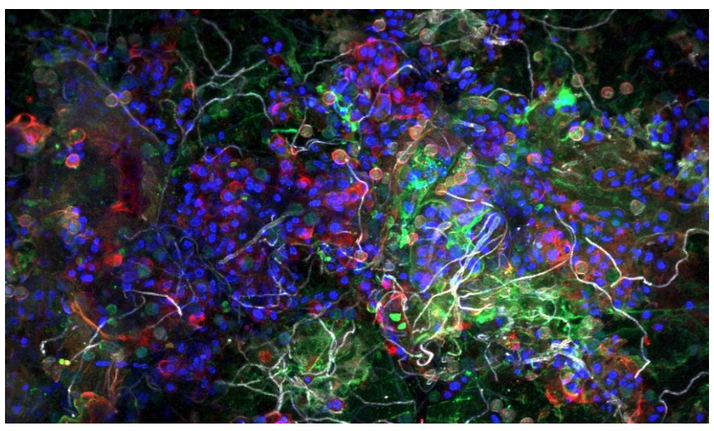
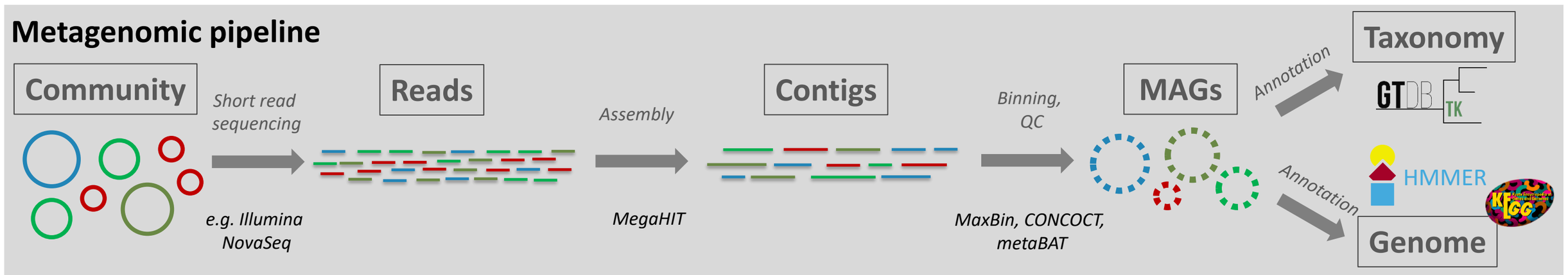
Multiple Stressor Ecology (mStress) working group
Department Ecotoxicology

# Molecular tools to assess microbial diversity and functions

## Background & Methods

Microbial freshwater communities (biofilms) host a diversity of phyla, such as microalgae (blue), bacteria/Archaea (green) and fungi (white) that share a habitat on submerged surfaces embedded in extracellular polymeric substance (red). Thanks to their metabolic diversity, biofilms serve several **key functions for the ecosystem** such as primary production, nutrient (re)cycling or the degradation of natural and anthropogenic chemicals.



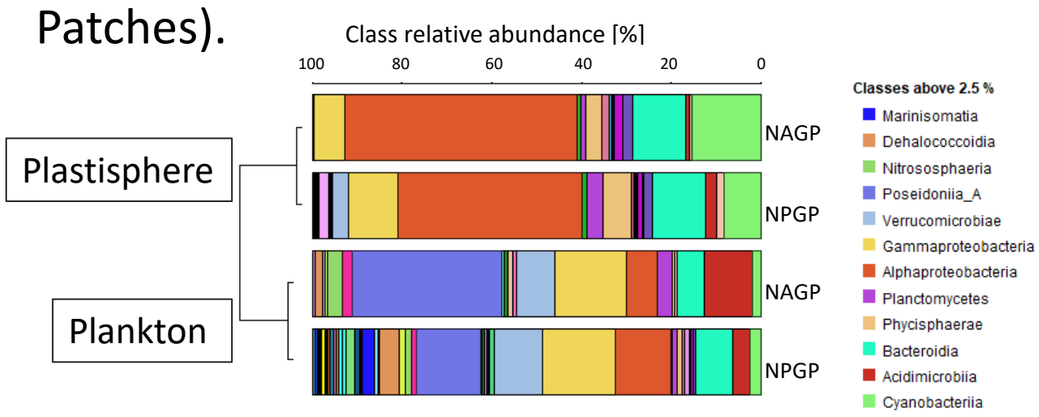



## Research: The plastisphere - a novel functional entity in the oligotrophic ocean

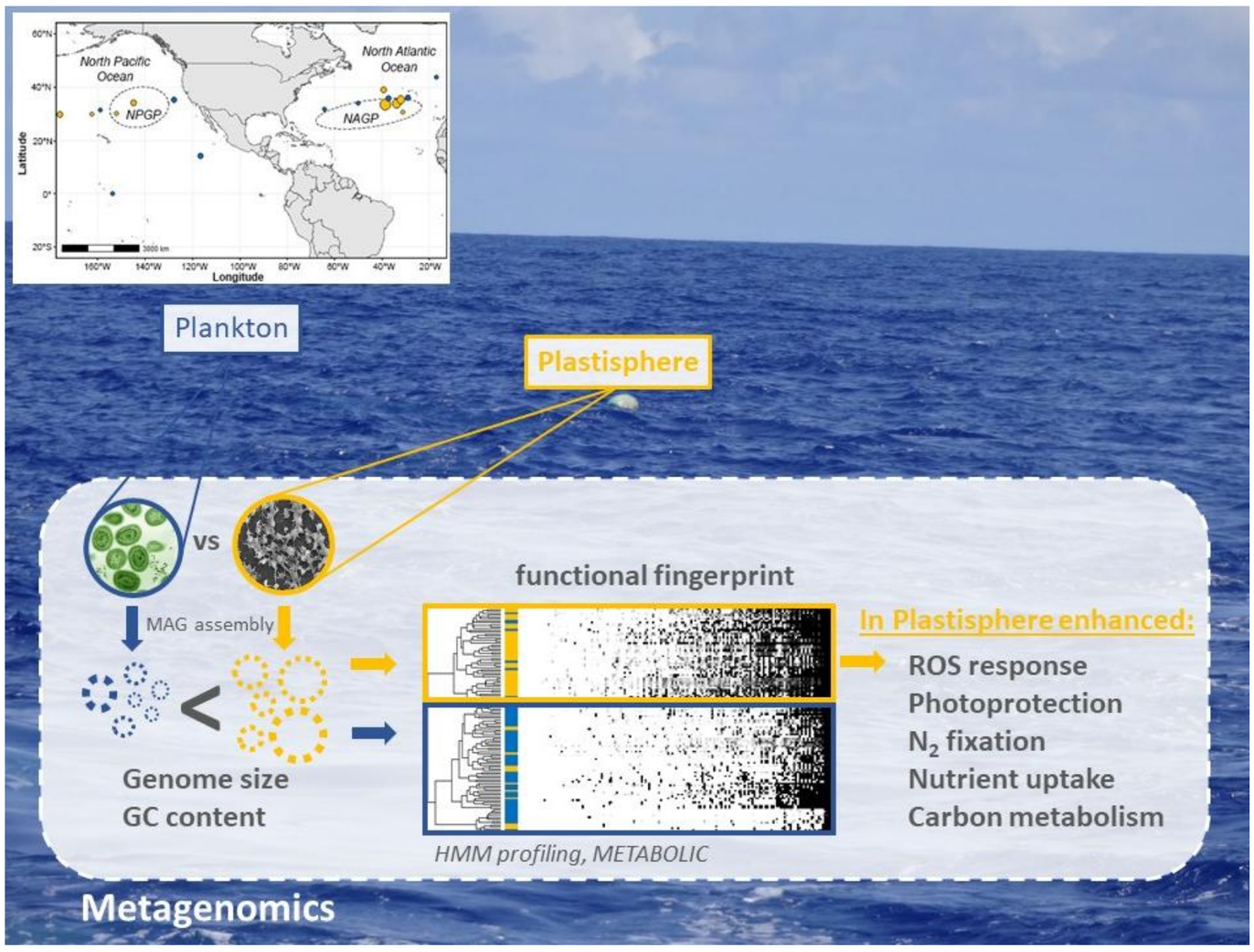
### Background

Trillions of plastic particles have accumulated in the oligotrophic ocean gyres, covered by microbial biofilms (=plastisphere).

We examined the **taxonomic** and **genomic differences between the plastisphere and the ambient plankton** in different gyres (North Atlantic & North Pacific Garbage Patches).




**Taxonomy**: The plastisphere is taxonomically distinct from the plankton, yet similar across oceans, indicating a high degree of functional redundancy.




**Metagenomics**: The plastisphere has bigger genomes, more nitrogen rich GC content, a distinct functional fingerprint and pathways enhanced, that allow the plastisphere to actively shape its nutrient supply, metabolize a broader range of substrates and benefit from high light conditions (e.g. engage in photoheterotrophy).

## People & Projects

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Federal Ministry of Education and Research
HELMHOLTZ Innovation
Funded by the European Union

## References


- **Lips, S., Schmitt-Jansen, M.**, & Borchert, E. (submitted). Metagenomic analyses of the plastisphere reveals a common functional potential across oceans
- Scales, B.S., Hassenrück, C., Moldaenke, L., Hassa, J., Rückert-Reed, **C., Rummel,** C., Völkner C., Rynek R., Busche T., Kalinowski J., Jahnke A**., Schmitt-Jansen, M**., Wendt-Potthoff K., Oberbeckmann S. (2024) Hunting for pigments in bacterial settlers of the Great Pacific Garbage Patch. Environmental Microbiology, 26(6), e16639. https://doi.org/10.1111/1462-2920.16639
- Tora, D., Hentschel, U., **Lips, S., Schmitt-Jansen, M**., Borchert, E. (2023) 16S rRNA gene sequence analysis of the microbial community on microplastic samples from the North Atlantic and Great Pacific Garbage Patches. Afr. J. Microbiol. Res. 17 (5), 123 – 138https://doi.org/10.5897/AJMR2022.9682
- **Lips, S., Larras, F., & Schmitt-Jansen, M.** (2022). Community metabolomics provides insights into mechanisms of pollution-induced community tolerance of periphyton. *Science of The Total Environment*, *824*, 153777. https://doi.org/10.1016/j.scitotenv.2022.153777

MANTRA

Begleitvorhaben zur Material-Hub-Initiative MaterialNeutral – einer Initiative des Bundesministeriums für Forschung, Technologie und Raumfahrt

# Daten zu innovativen Materialien für Nachhaltigkeit und Transfer

## Aufgaben und Ziele von MANTRA

**Allgemein:**
- Begleitvorhaben der Material-Hub-Initiative “MaterialNeutral”
- Betrachtung der Sicherheit von innovativen Materialien
- Nachhaltigkeitsbetrachtung innovativer Materialien
- Kommunikation für Projekte und Öffentlichkeit über unsere Wissenbasis https://materialneutral.info

**Für die geförderten Projekte:**
- Vernetzung (Workshops und Statuskonferenzen)
- Öffentlichkeitsarbeit
- Entwicklung geeigneter Nachhaltigkeitsindikatoren
- Industrie- und Praxistransfer

## Materialien für mehr Prozesseffizienz

In Modul 1 unterstützt MANTRA 15 F&E Projekte, die innovative Materialien für **Katalyse** und/oder Membranen entwickeln:

**AmmoCatCoat**
**CARBO-DIOL 2.0**
**DisCO2very**
**ReLaKiKat**
**ReMAOlution**
**PerDemand**
**MyrRems**
**MeGaPho**

**BETA**
**COBRA**
**i-KaMeRa**
**UltraPd**

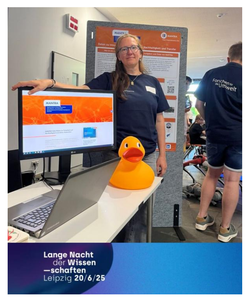
Helios
NanoKom
KeraRes

→ https://materialneutral.info/forschung/projekte/

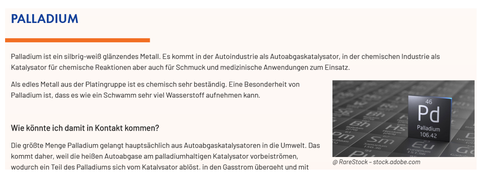
## Unsere Aktivitäten

### Entwicklung von Nachhaltigkeitsindikatoren
- 1. Workshop mit den Projekten (April 2025, Berlin)
- Clustern & Priorisieren der Workshop-Ergebnisse
- Definition von Auswahlkriterien für Indikatoren
- Erstellung eines Basis-Sets an Indikatoren, optionale Erweiterungen
- Geplant: 2. Workshop (virtuell, 1. Quartal 2026)


### LinkedIn
- Vorstellung aller MANTRA-Teammitglieder
- Vorstellung aller Projekte
- Regelmäßige Updates
- Geplant: posts zu FAQs + Projekten aus Modul 2









Digitalisierung und Forschungsdaten-Management

Die Digitalisierung Digitalisierung beschreibt einen Prozess, bei dem man analoge Informationen und Prozesse in eine digitale Form überführt. Das bedeutet, […]

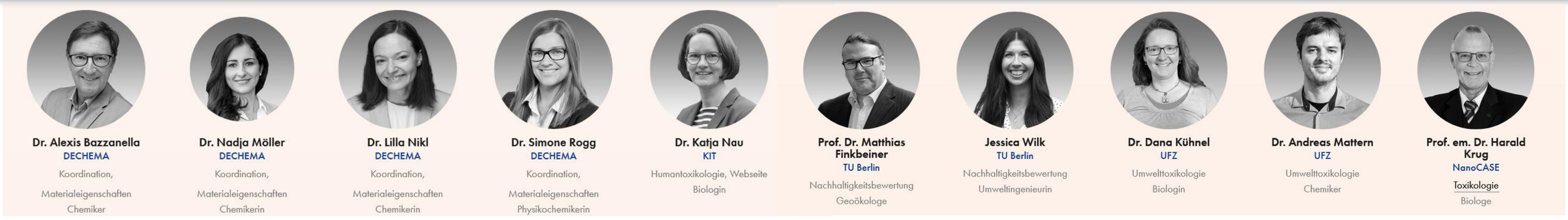
WEITERLESEN

### Neue Inhalte Webseite
- Grundlagentexte Katalyse, Membranen und Nachhaltigkeit
- FAQs & regelmäßige News
- Digitalisierung
- Neue Materialien

### Tool für LCA und techno-ökonomische Analysen aus Prozesssimulationen
- Webinar zur Vorstellung des Tools
- Umsetzungsprojekte (in Planung)

### Vernetzung MaterialDigital
- Gemeinsame Workshops zum PMD Datenportal (virtuell, März und Juni 2025)
- Sprechstunden für Projekte zur Beratung

## Das Team hinter MANTRA



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**Wir vereinen Fachwissen aus Biologie, Toxikologie, Chemie, Material- und Umweltwissenschaften, sowie der Nachhaltigkeitsbewertung.**

Gefördert durch:
Bundesministerium für Forschung, Technologie und Raumfahrt
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projekt mantra

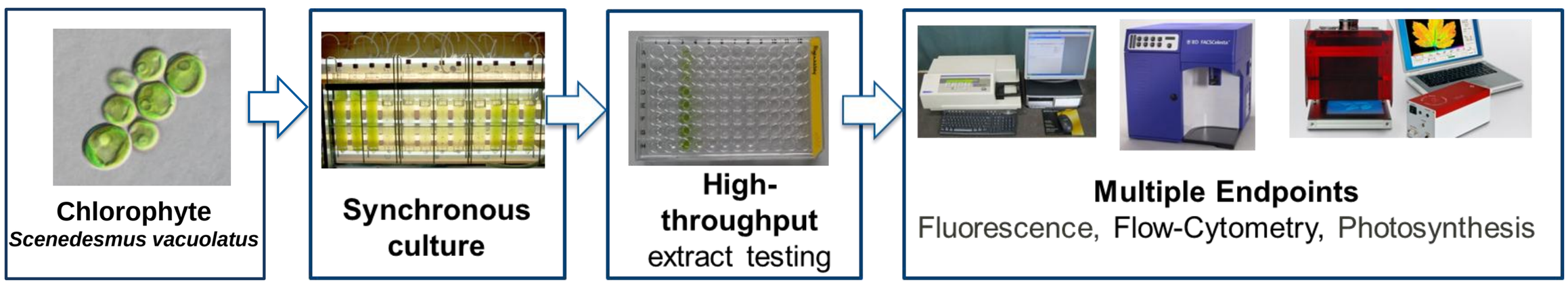
Multiple Stressor Ecology (mStress) working group
Department Ecotoxicology

# A diagnostic high-throughput algae test for effect-based monitoring

## Background & Method

For **Effect-based monitoring** the assessment of the phytotoxic potential of environmental contaminants is essential. We developed and apply a multi-endpoint high-throughput method applicable for single substances, environmental samples (water and extracts) and technical mixtures like plastic leachates.

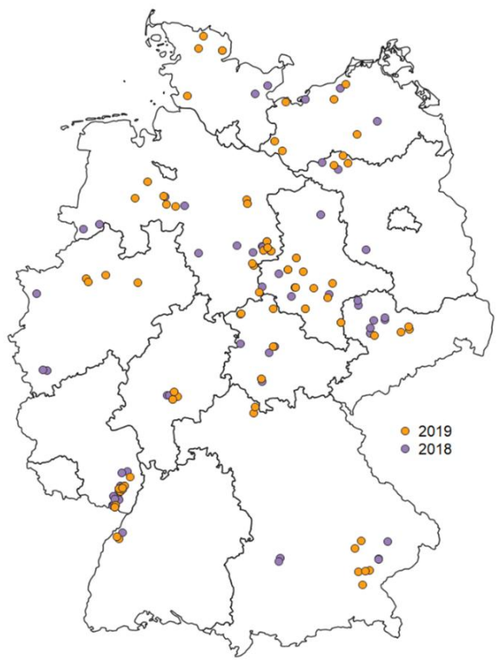
The **Diagnostic Algae Test (DAT)** used synchronized cultures (24h) of the Chlorophyte *Scenedesmus vacuolatus* in a 96 well-plate format to derive dose-dependent responses on apical (growth) and physiological endpoints (photosynthesis).



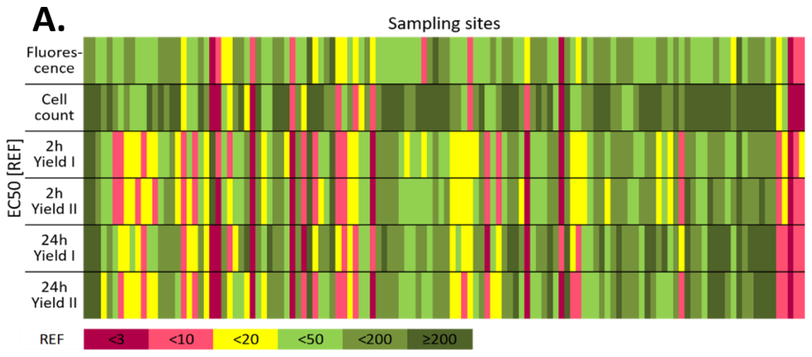

## Case study: Assessing the phytotoxicity of agricultural run-off in small streams - the KGM (Kleingewässermonitoring) - project

**Aim:** Analyze the potential and effects of agricultural run-off on the Biological Quality Element 'Phyto' in Germany
**Hypothesis:** Agricultural streams are exposed to high toxicity resulting in shift in biofilm (diatom) communities

## Key Results:

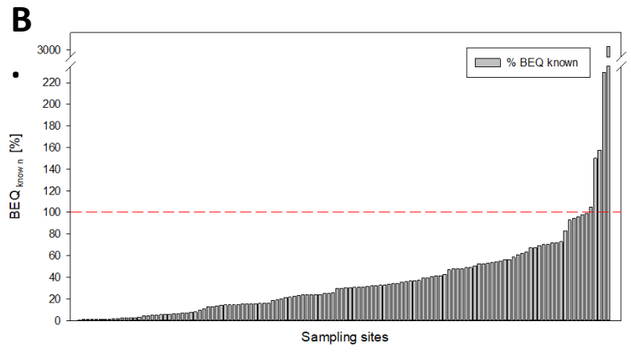



**LAB-approach: Effect based monitoring** → **Field-approach: Community structure of biofilms**

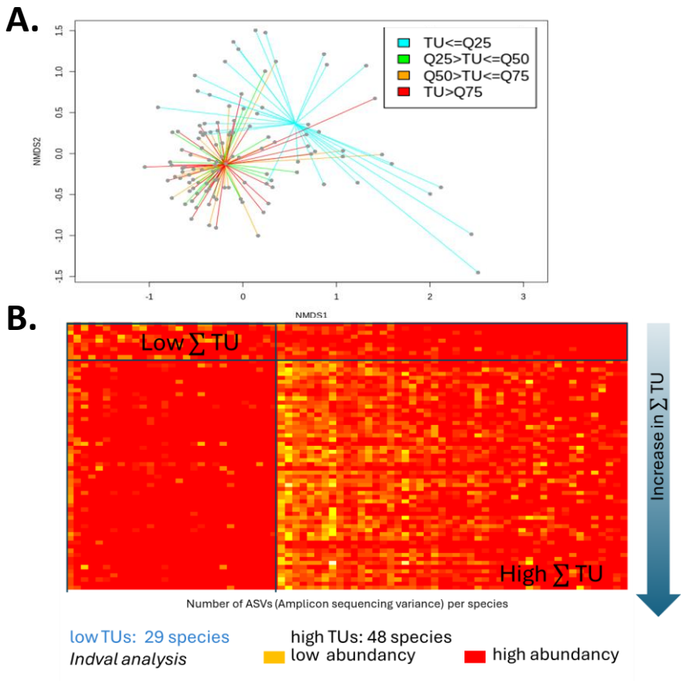



biomass-related endpoints

photosynthesis-related endpoints




| $BEQ_{known}$ [%] | % of Sites |
|---|---|
| 0-33 % | 58 |
| 33-66 % | 27 |
| 66-100 % | 11 |
| >100 % | 4 |
| 30 % | median |




**Study design:**

- 64 sites across Germany
- 116 samples (water extract)
- Metabarcoding of 60 biofilms

**A: DAT** revealed a high toxic potential for alga esp. for photosynthesis ➔ dominance of herbicides
**B: Iceberg modelling** could explain 30% of effect based on 109 pesticides

**A: NMDS based on 4008 diatom** ASVs reveals clear separation of sites with low TUs
**B: IndVal** analysis reveals 48 diatom species associated with high TUs

➔ **High phytotoxicity** of agricultural run-off but high portion of effects remains unexplained
➔ **High risk for primary producers** in agricultural streams with shifts in communities in the BQE 'Phyto'

## People & Projects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KgM Kleingewässer Monitoring · Umwelt Bundesamt · P-LEACH · HELMHOLTZ Innovation · JPI OCEANS WEATHER-MIC · Bundesministerium für Bildung und Forschung

## References


- Rummel C.D., Schäfer H., Arp HP., Jahnke A., **Schmitt-Jansen M.** 2022 Effects of leachates from UV-weathered microplastic on the microalgae *Scenedesmus vacuolatus*. Anal Bioanal Chem. 414 (4), 1469 – 1479.
- Liess, M., Liebmann, L., Lück, M., Vormeier, P., Weisner, O., Foit, K., Knillmann, S., Schäfer, R.B., Schulze, T., Krauss, M., Brack, W., Reemtsma, T., Halbach, K., Link, M., Schreiner, V.C., Schneeweiss, A., Möder, M., Weitere, M., Kaske O., von Tümpling, W., Gunold, R., Ulrich, N., Paschke, A., Schüürmann, G., **Schmitt-Jansen, M., Küster, E.**, Borchardt, D. 2022 Umsetzung des Nationalen Aktionsplans zur nachhaltigen Anwendung von Pflanzenschutzmitteln (NAP) – Pilotstudie zur Ermittlung der Belastung von Kleingewässern in der Agrarlandschaft mit Pflanzenschutzmittel-Rückständen. Abschlussbericht, Forschungskennzahl 3717 63 403 0 Texte Umweltbundesamt 07/2022, 319 S.
- Schmidt, M., **Aulhorn, S.**, Latif, A.A., Krauss, **M., Schmitt-Jansen**, M., Breite, D., **Küster, E.,** Schulze, A. (2025): Photocatalytic membrane treatment of antibiotics: combined chemical and toxicological evaluation of effectiveness Front. Env. Sci. Eng. 19 (12), art. 163 10.1007/s11783-025-2083-7

Multiple Stressor Ecology (mStress) working group
Department Ecotoxicology

# Periphyton responses under a Multiple Stressor Framework

## Background & Methods

Freshwater ecosystems face **combined stress** like antibiotics pollution and heat waves. **Periphyton**, a key microbial biofilm sustaining aquatic ecosystem functions like primary production, is vulnerable to these pressures. Functional omics, dose–response modelling and the **Stress-Induced Community Tolerance (SICT)** approach are combined to link molecular stress traits with phenotypic responses and to define resilience thresholds under multiple stressors.

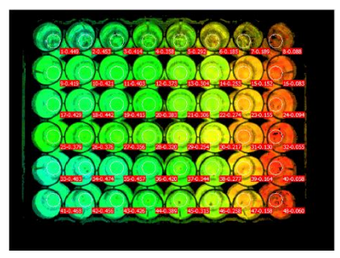
### Field sampling

### Multiple stressor exposure in microcosms

### Multi-Endpoint Effect assessment

➢ **SICT endpoint (Stress-induced community tolerance)**

- Photosynthesis activity
- Protein synthesis and gross primary productivity



➢ **Structural and functional changes analysis**

- Community composition
- Antibiotic resistance genes (ARGs) quantification
- Metabolic pathways (metabolomics)
- Pigments analysis

## Research: Combined effects of heat waves and antibiotic exposure on periphyton

**Research Question:** Do climate-related stressors (heat) alter the sensitivity of periphyton communities?

**Hypothesis:** During long-term exposure (4 weeks), realistic **heat waves** (from 20 to 25 °C) alter periphyton tolerance to **antibiotics (Erythromycin)**, affecting the functions and structure of these microbial communities

### Key Result

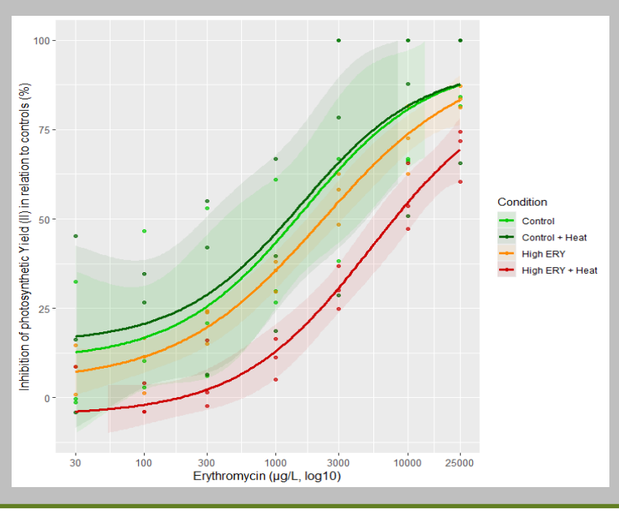
- Assessment of periphyton photosynthetic activity after the long-term exposure experiment, during a short-term re-exposure (24h) along a long-gradient design of Erythromycin

➔ **induced community tolerance after combined exposure to heat and antibiotics**

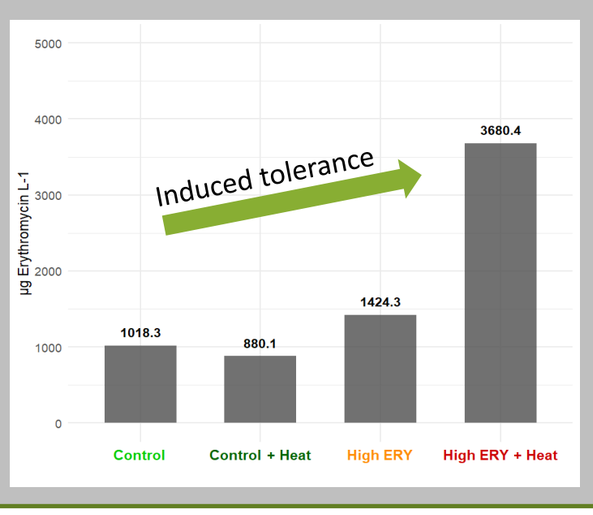
Conditions :

**- High ERY** = 100 µg/L Erythromycin exposure for 4 weeks

**- Heat =** 25°C water temperature for 2 weeks




**Short-term responses** (24 h) to Erythromycin of periphyton pre-exposed to heat and Erythromycin, individually and in combination




**SICT** (Stress-induced community tolerance): **EC50 values** of inhibition of photosynthetic activity of communities differing in pre-exposure to Erythromycin and heat

## People & Projects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PHARM - ERA
Funded by the European Union
CLIMSHIFT
Funded by DFG Deutsche Forschungsgemeinschaft German Research Foundation


## Publications


- Rogue H., Miège C., Bonnineau C., Daval A., Depret G., **Susset L**., Tardy V., Lyautey E., Devers M., Pesce S. (2025) Sulfamethazine biodegradation in sediments is driven by chronic exposure concentrations. Ecotoxicology and Environmental Safety, 303, 118785, https://doi.org/10.1016/j.ecoenv.2025.118785
- Vijayaraj, V., Laviale, M., Allen, J., Amoussou, N., Hilt, S., Hölker, F., Kipferler, N., Leflaive, J., López Moreira M., G.A., **Polst, B., Schmitt-Jansen, M**., Stibor, H., Gross, E.M. (2022): Multiple-stressor exposure of aquatic food webs: nitrate and warming modulate the effect of pesticides. Water Res. 216, art. 118325 https://doi.org/10.1016/j.watres.2022.118325
- **Polst B.H**., Hilt S., Stibor H., Hölker F., Allen J., Vijayaraj V., Kipferler N., Leflaive J., Gross E.M., **Schmitt-Jansen M**. (2022) Warming lowers critical thresholds for multiple stressor–induced shifts between aquatic primary producers. Sci. Total Environ 838 art. 156511 https://doi.org/10.1016/j.scitotenv.2022.156511

Multiple Stressor Ecology (mStress) working group
Department Ecotoxicology

# From the laboratory to the field – the *Lymnaea stagnalis* test battery for assessing acute, chronic and transgenerational toxicity

## Background & Methods

***Lymnaea stagnalis*** is a common freshwater snail, serving the ecosystem as grazer and food source. The snail has become an attractive model organism in developmental biology and (eco)toxicology. It is easy to breed, has a high reproduction rate, and, as it is not subject to animal welfare regulations, the various life stages (embryo, juvenile, adult) offer the opportunity to perform acute and chronic toxicity assays up to transgenerational tests.

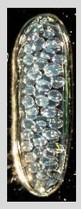
**Life stages:**

**Adult:** Reproduction test

- Chronic toxicity assessment (OECD 243)
- Duration: 28 days

**Endpoints:**

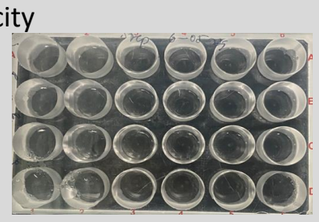
- Feeding rate
- Behavior
- Reproduction
- Mortality



**Transgenerational test**

**Embryo:** Acute toxicity test
- the Snail Embryo Test (SNET)

- Screening tool for acute toxicity
- Duration: 14 days



**Endpoints:**

- Mortality
- Sublethal effects (malformations, pigmentation, shell size, hatching, heart rate)

Field monitoring

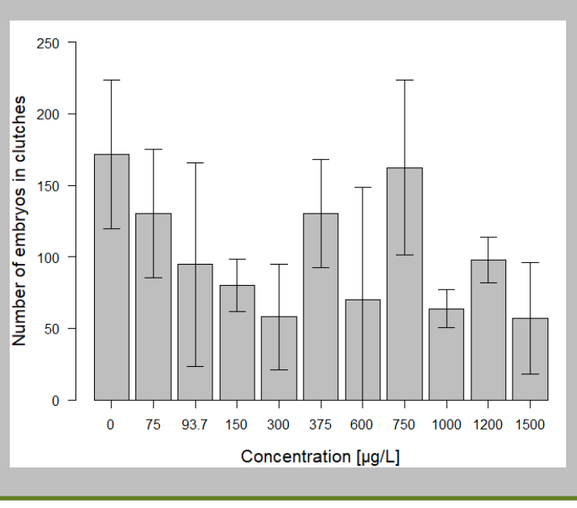

MOBICOS or *in-situ* caging experiments at point sources, e.g. Waste Water Treatment Plants (Silsted, Harz)
(in planning)

**Case study: From Plastic Additives to Ecological Hazard:** The effects of Tetrabromobisphenol A (TBBPA) on *Lymnaea stagnalis*

## Introduction

- TBBPA is a  widely used flame retardant, prominent substitute of Polybrominated Diphenyl Esters (PBDEs)
- high log KOW(5.9) -> persistent, bioaccumulative and toxic substance ?
- ecotoxicological effect is largely unknown

## Key Results

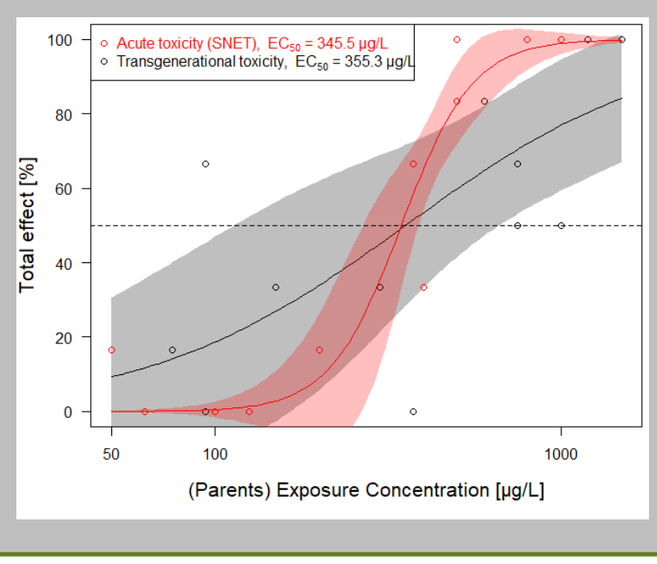



### » altered reproduction

TBBPA exposure reduces number of offsprings. At 375 and 750 µg $L^{-1}$ an induction was observed.

### toxic parents «

Offsprings of exposed adults and acute exposed embryos in the SNET from unexposed adults show similar toxicity.




## Summary & Conclusion

- TBBPA affected reprodution and behaviour (data not shown) and toxicity was observed in the next generation (F1) of exposed adults.
- Embryo‘s from unexposed adults (analysed by SNET) showed similar toxicity after acute exposure to TBBPA.
- Transfer of TBBPA via generations and uptake from the exposure solution may result in similar exposome in embryo‘s.

  -> Chemical analysis of internal concentration (Alexander Böhme, Dep. Expo currently running).

## People & Projects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