



NORMAN WWTP Project

European case study on wastewater treatment plant effluents



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Leipzig



Objectives

- Identify relations between measurable contamination and effects in biotests
- Identify typical drivers of effects
- Identify typical chemical and effect patterns



Sampling campaign – 57 LVSPE WWTP effluent samples

Jörg Ahlheim (WANA/UFZ)



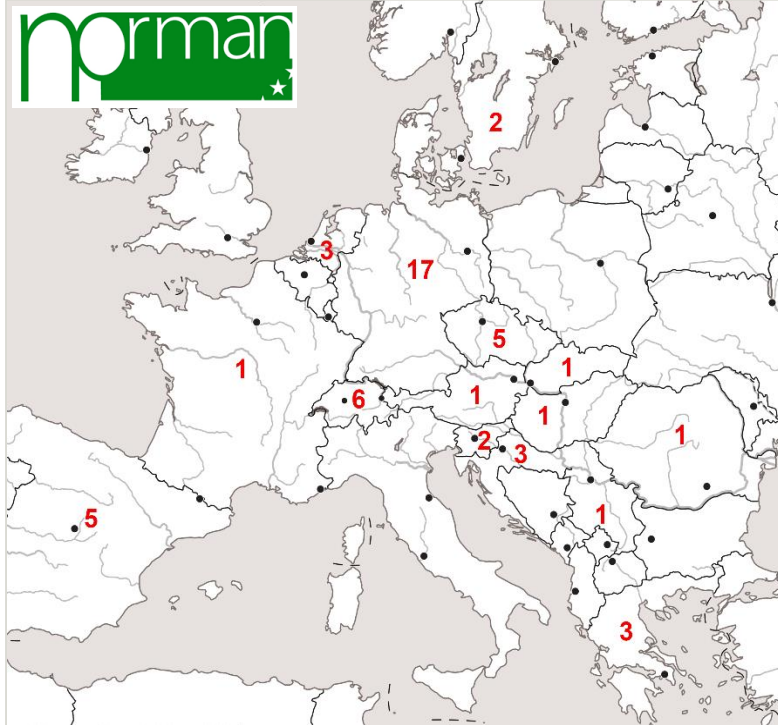
Foto by Jörg Ahlheim



Foto by USDA (www.wikipedia.org)

General information

- Large Volume Solid Phase Extraction (LVSPE)
- 52 European WWTPs (5 sampled 2x)
 - **57 LVSPE effluent samples**



15 Countries

- Germany (17)
- Switzerland (6)
- Czech Republic (5)
- Spain (5)
- Croatia (3)
- Greece (3)
- Netherlands (3)
- Slovenia (2)
- Sweden (2)
- Austria (1)
- France (1)
- Hungary (1)
- Romania (1)
- Serbia (1)
- Slovakia (1)

Chemical target screening

- ✓ **Wide-scope target screening (~500 compounds)** (WANA, UFZ)
- ✓ **Screening of 80 steroids and phenols** (WANA, UFZ)

Effect screening – *in vivo*

- ✓ **Algae bioassay I** (ÖZ/EAWAG)
- (✓) **Algae bioassay II** (BIOTOX, UFZ)
- (✓) **Daphnia bioassay** (BIOTOX, UFZ)

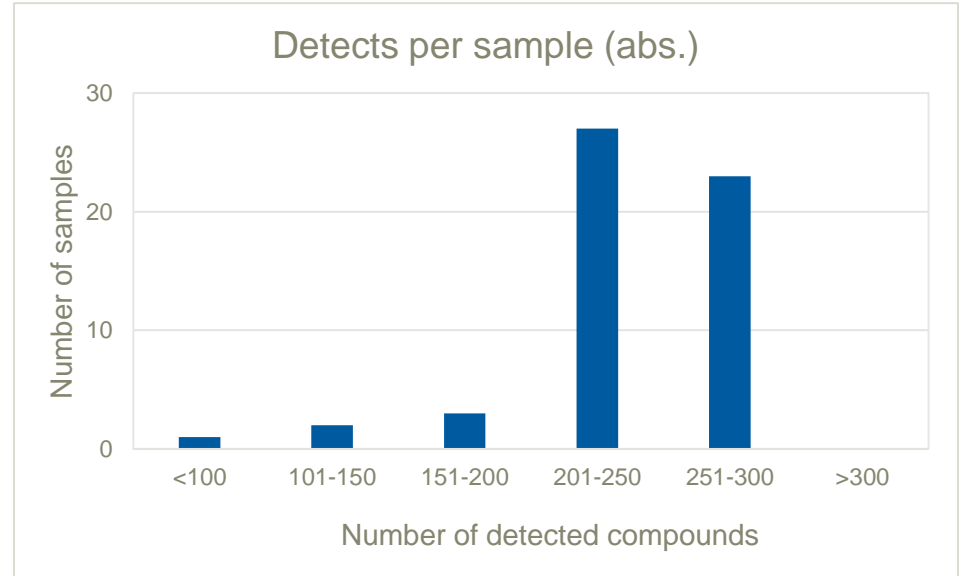
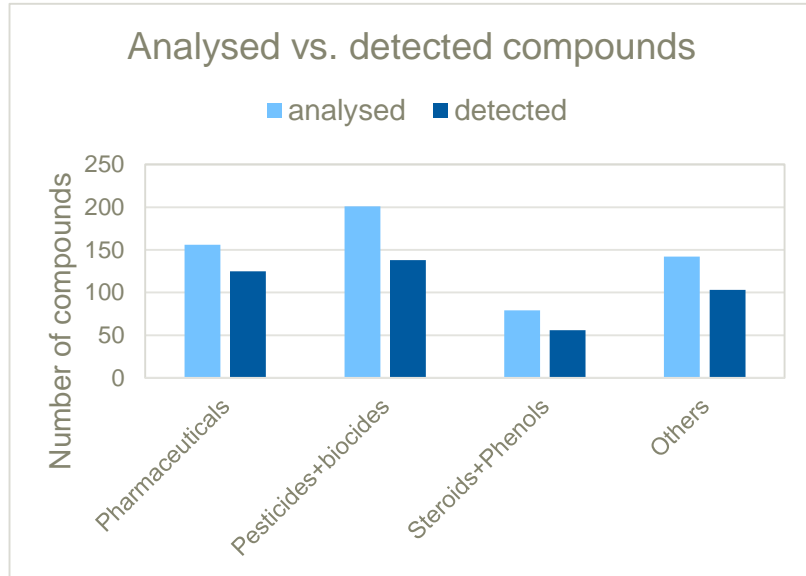
Effect screening – *in vitro*

- ✓ **GeneBLazer assays: ER α , GR, AR, PR** (CELLTOX, UFZ)
- (✓) **CALUX assays: ER α , Anti-AR** (RWTH Aachen)
- ✓ **Planar Yeast Estrogen Screen (p-YES)** (BfG)



Detected compounds

S. Finckh and W. Leekitratanapisan (WANA/UFZ)



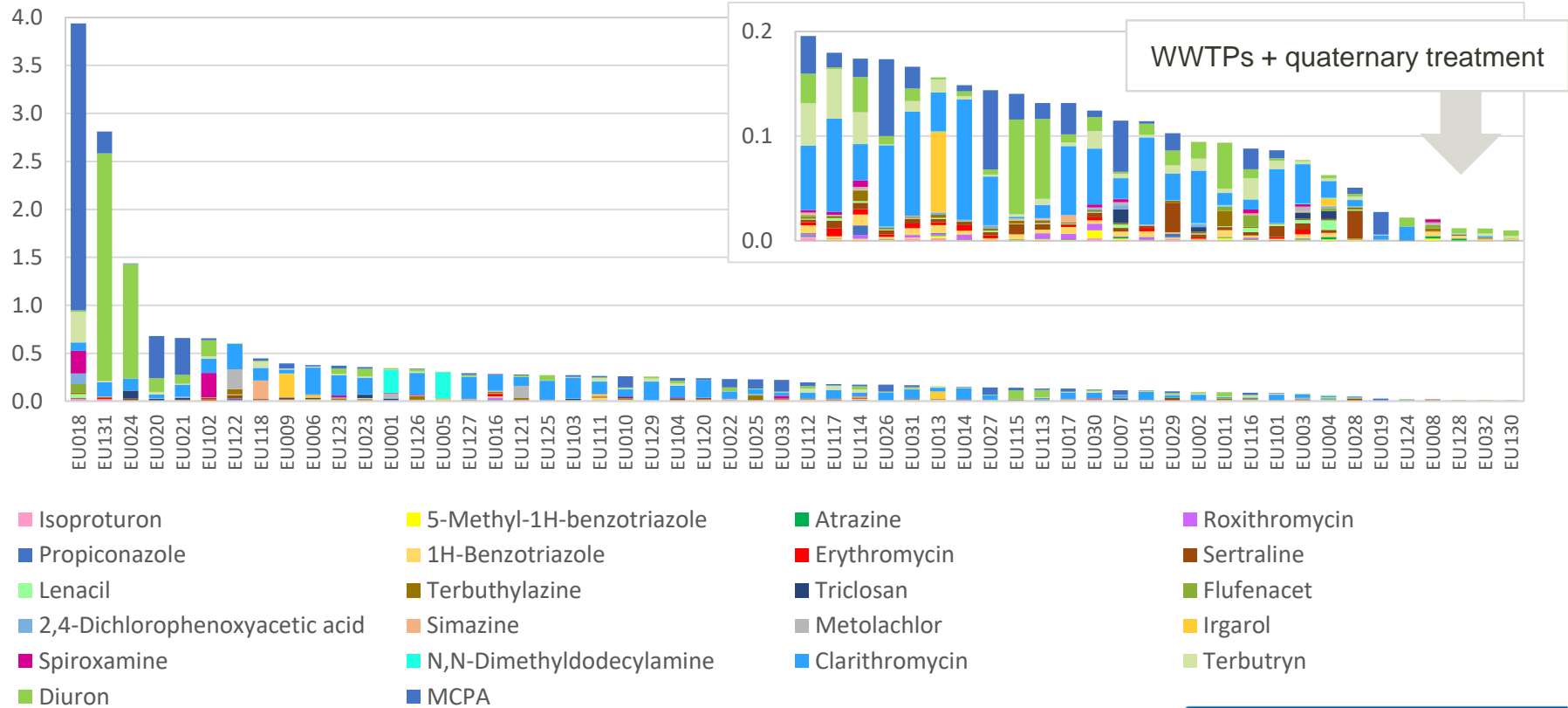
➤ Analysed compounds: 578

➤ Detected compounds: 422

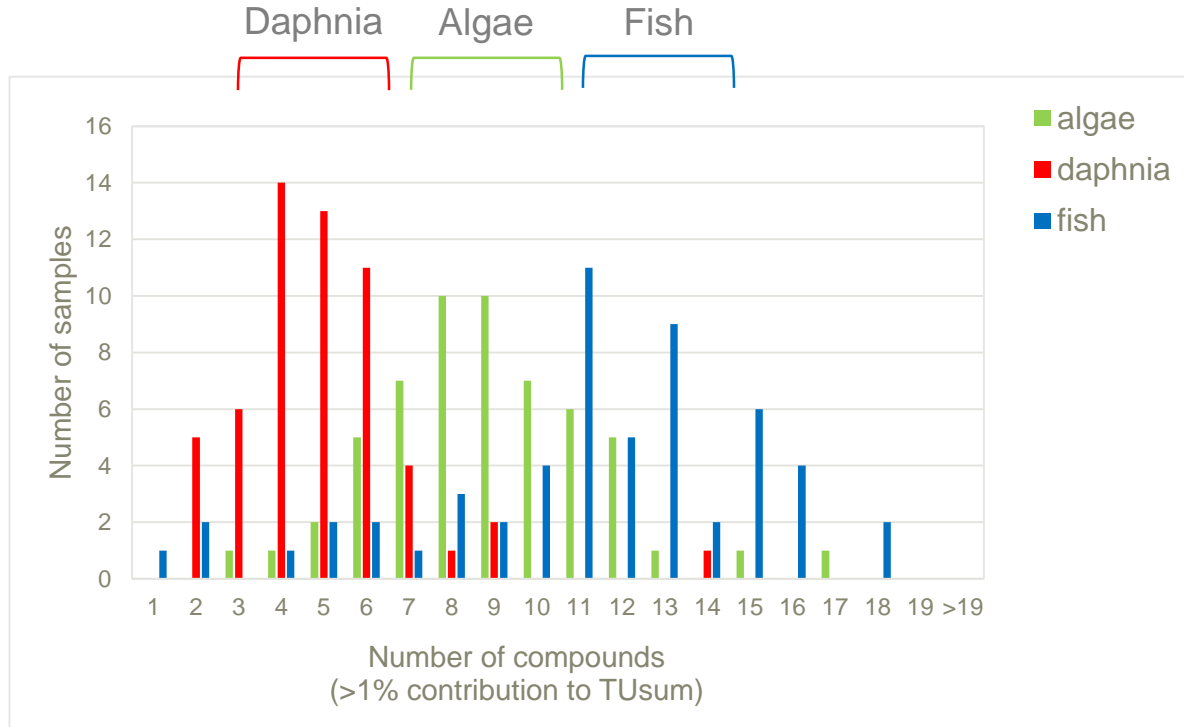
➤ Detected compounds per sample: 200-300

➤ Many target compounds frequently detected

Risk assessment – Algae toxicity (TU approach)



TUsum: How many compounds contribute >1% ?



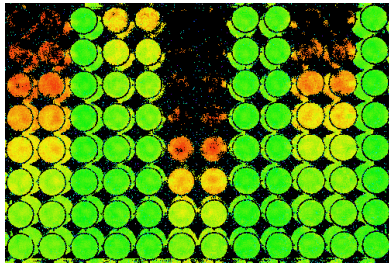
Drivers of Toxicity

- **Daphnia**
 - 4-6 compounds
- **Algae**
 - 8-10 compounds
- **Fish**
 - 11-13 compounds

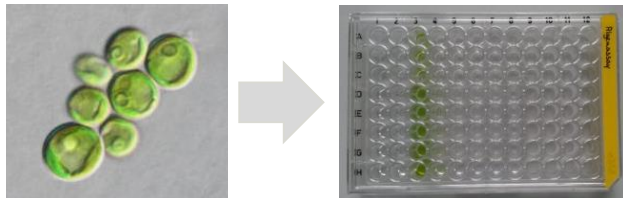
Algae bioassays – PSII and growth

E. Vermeirssen (ÖZ/EAWAG), M. Schmitt-Jansen (BIOTOX/UFZ)

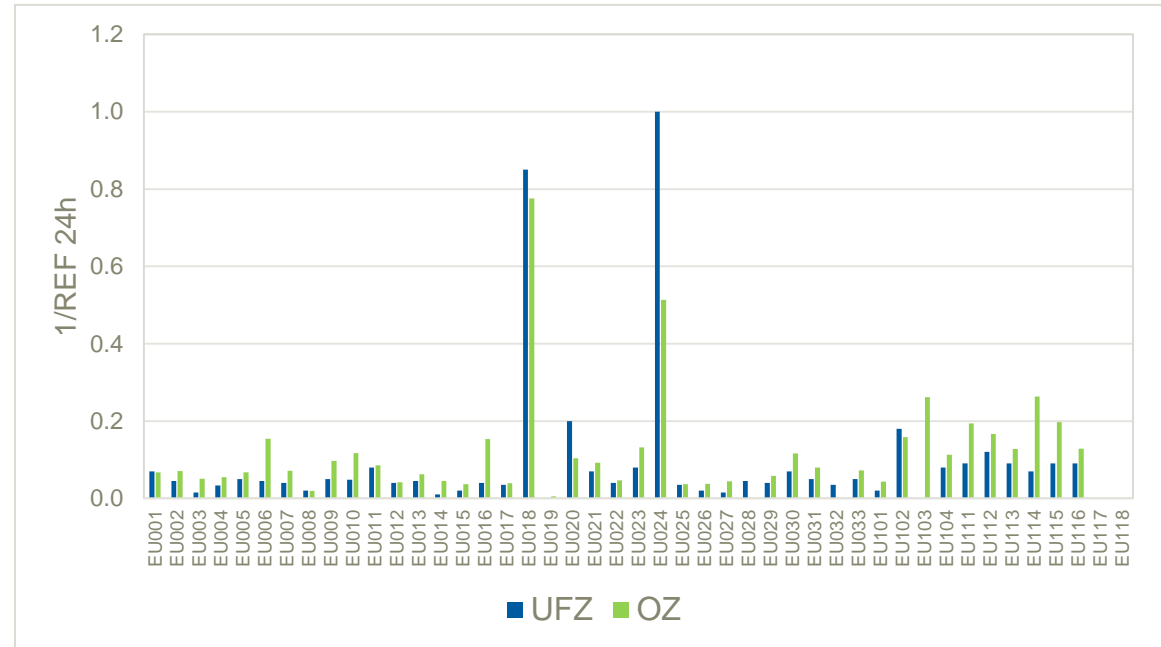
Pseudokirchneriella subcapitata



Scenedesmus vacuolatus



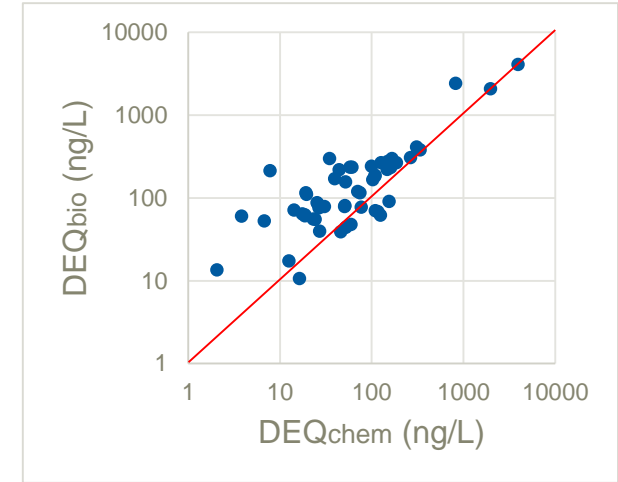
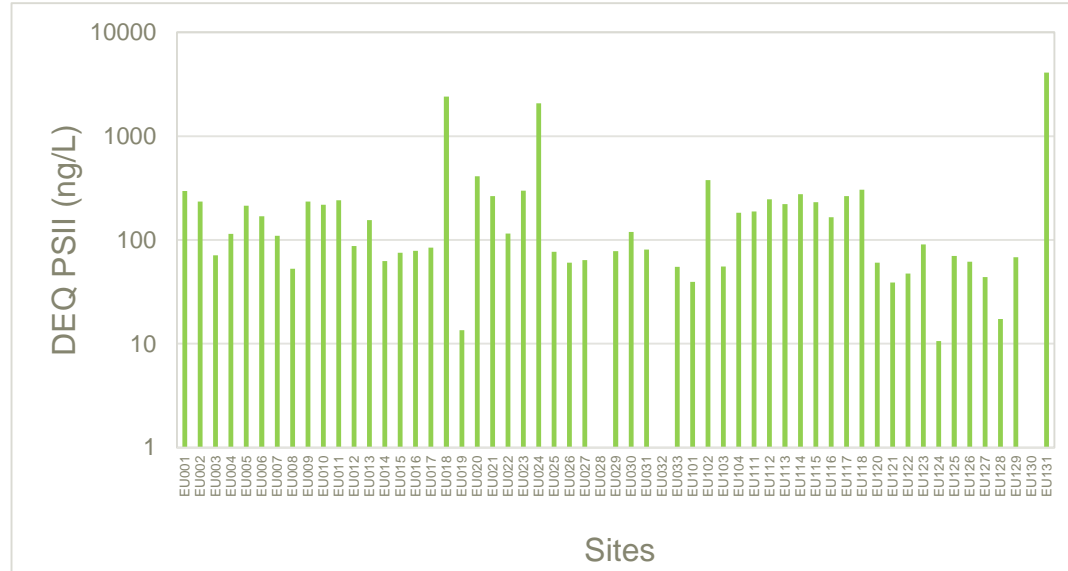
PSII inhibition – REF = Relative Enrichment Factor



Algae toxicity – chemical vs. effect data

E. Vermeirssen (ÖZ/EAWAG), S. Finckh (WANA/UFZ)

PSII inhibition (algae bioassay I) – DEQ = Diuron Equivalents

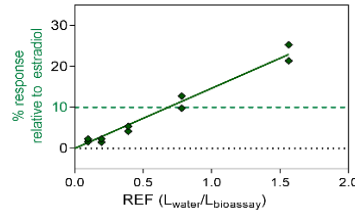


Diuron	Terbutylazine
Isoproturon	Terbutryn
Chlorotoluron	Atrazine
Metribuzin	Irgarol
Metamitron	Simazine
Chloridazon	Lenacil
Linuron	

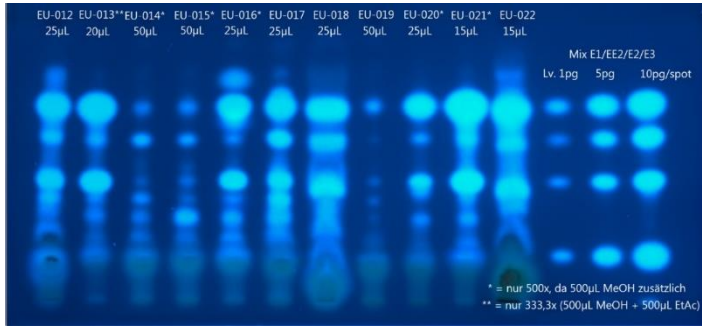
Estrogenicity – GeneBLAzer ER and pYES

S. Buchinger (BfG), W. Leekitratanapisan (WANA/UFZ), B. Escher (CELLTOX/UFZ)

GeneBLAzer ER



Planar Yeast Estrogen Screen (pYES)

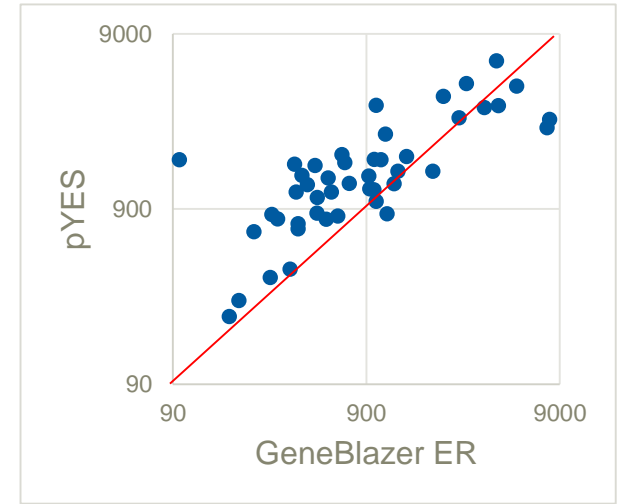


E1
 EE2
 E2
 E3

Samples

References

Estradiol Equivalents EEQ (pg_{E2}/L)



Next steps...

- 1st publication in progress (chemical target screening)
- December 2020: Collect all final data
- January 2021: Virtual project meeting
- Discussion on further publications:
 - “*in-vivo* publication”
 - “*in-vitro* publication”

Thank you!

