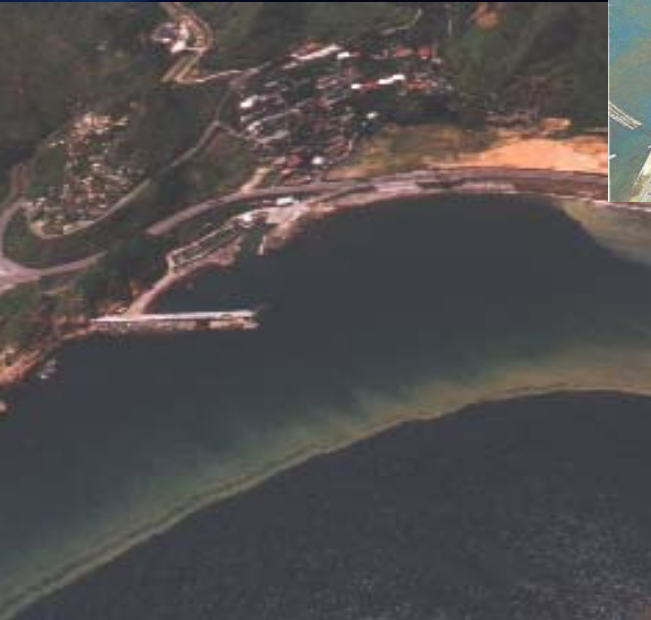
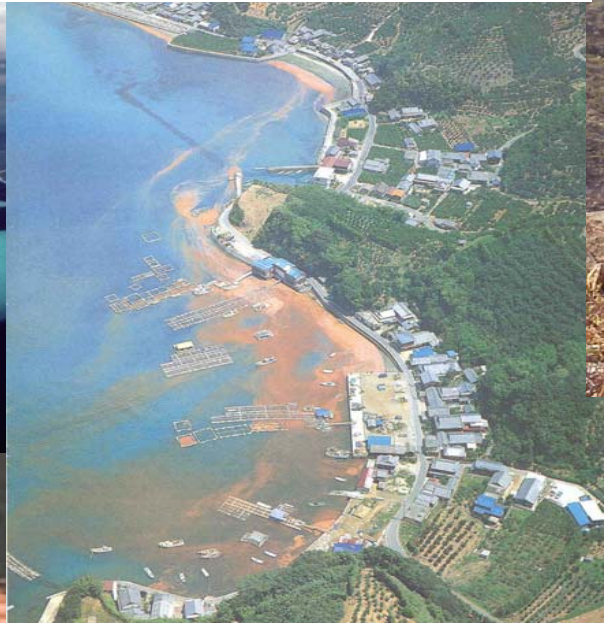


Chemical Ecology of Toxic Algae

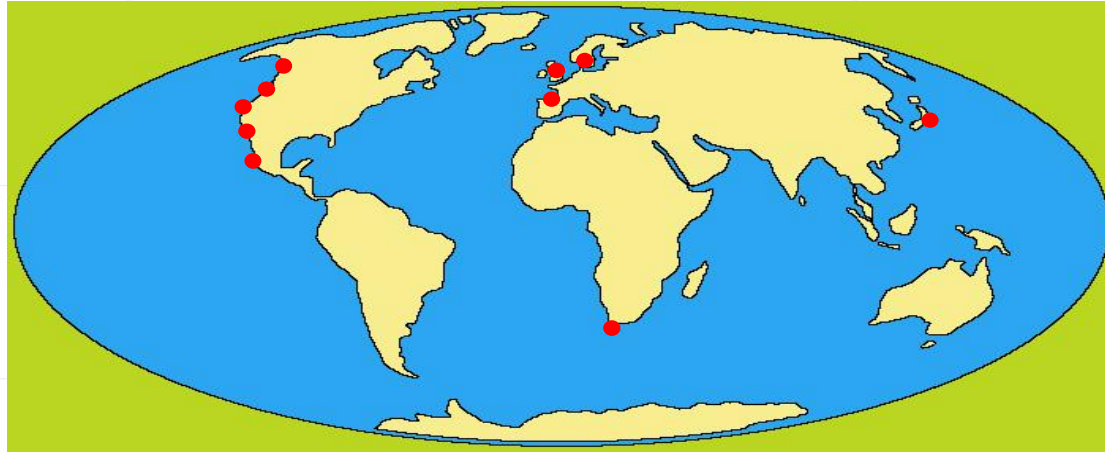
Bernd Krock, Urban Tillmann, Uwe John, Sára Beszteri,
Chishimba M. Kantu, Allan D. Cembella

Toxic Algal Blooms

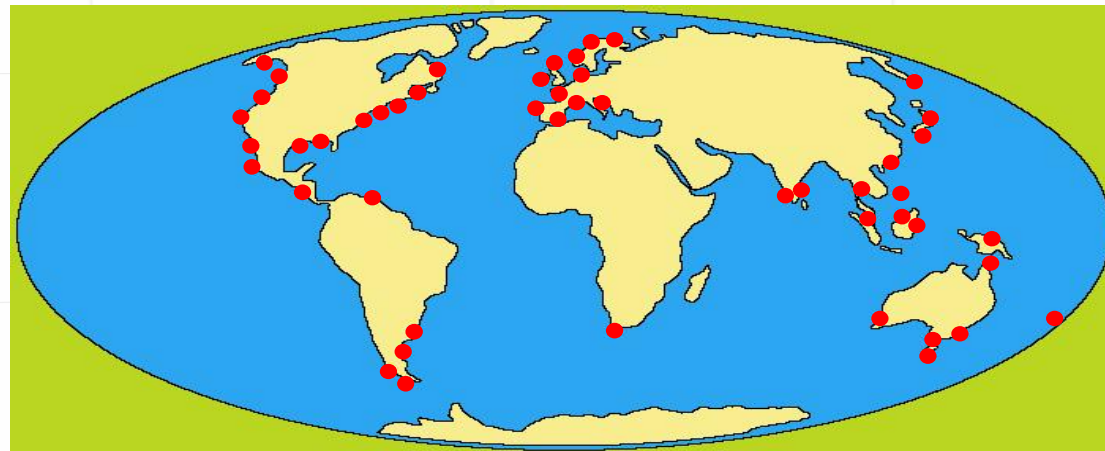


Distribution of Paralytic Shellfish Poisoning events

1970



2005



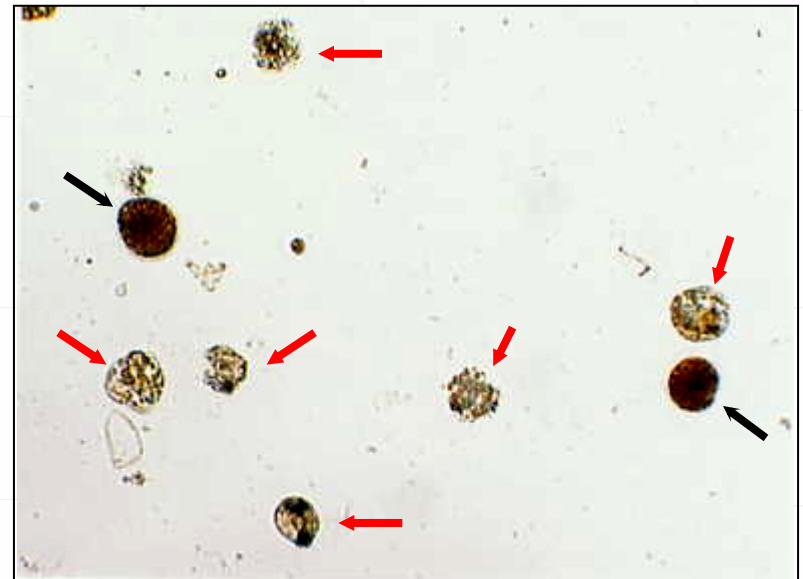
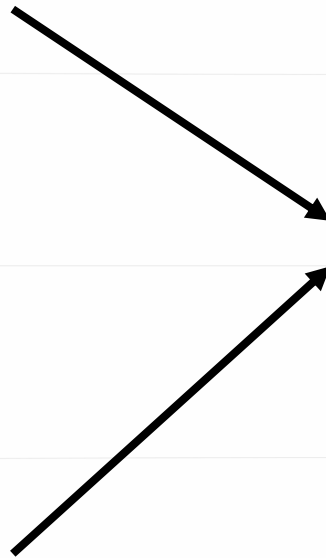
Lytic Effect of *Alexandrium*



Oxyrrhis marina



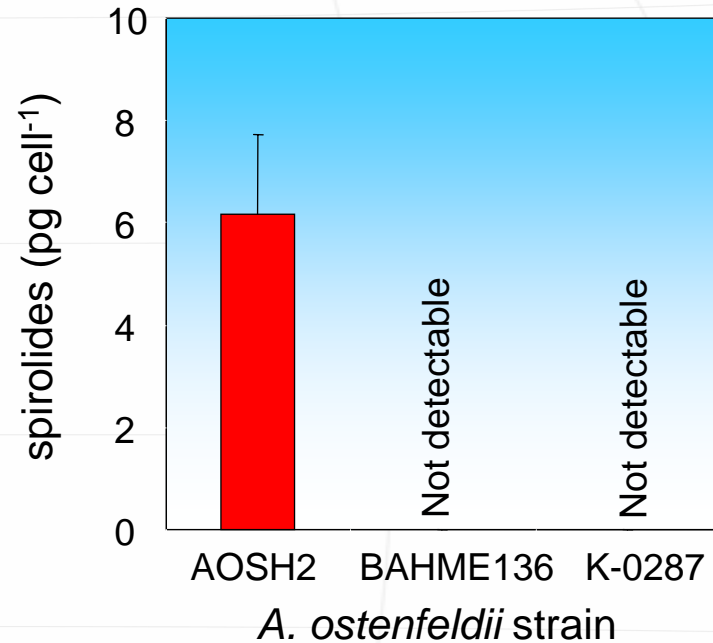
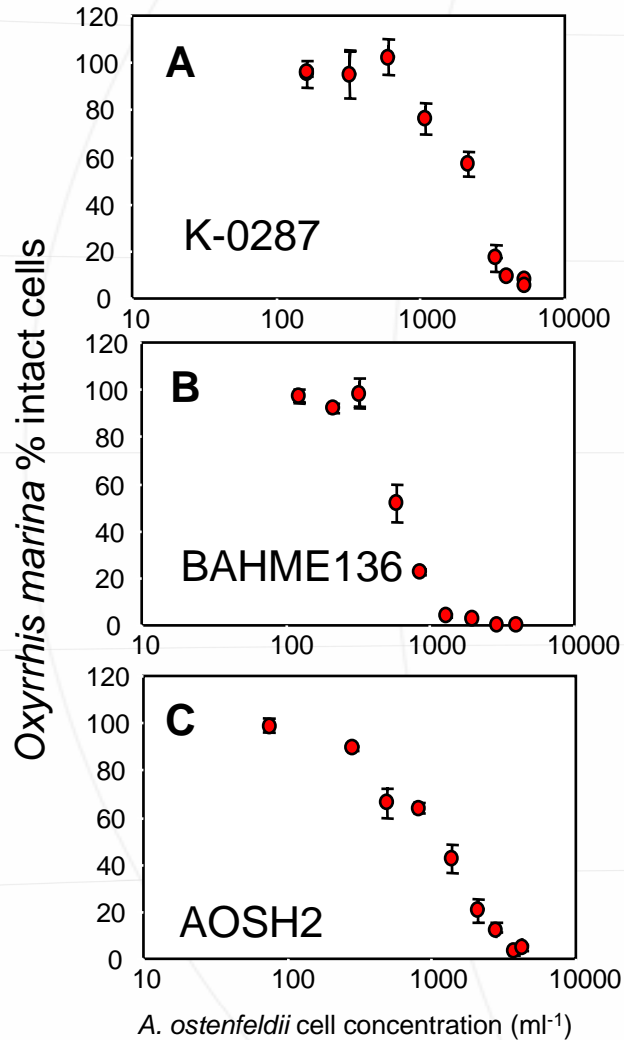
Alexandrium tamarense



Lytic Effect of *Alexandrium* shown with *Oxyrrhis marina*.

Black arrows: *Alexandrium*
Red arrows: Reminders of *Oxyrrhis*

Lytic Effect of *A. ostenfeldii*



Allelochemical potency is not related to spiroside production

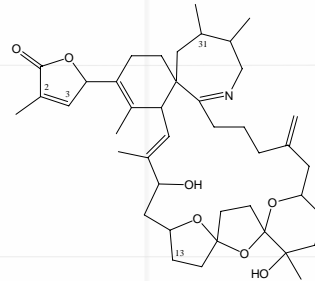
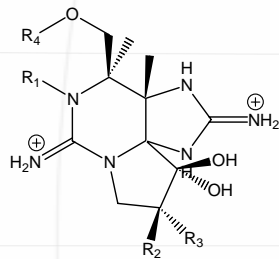
Organism Chemical Interaction Ecological Function

Alexandrium tamarense

PSP-Toxins

Spirolides

Alexandrium minutum



?

Alexandrium ostenfeldii

Alexandrium tamarense

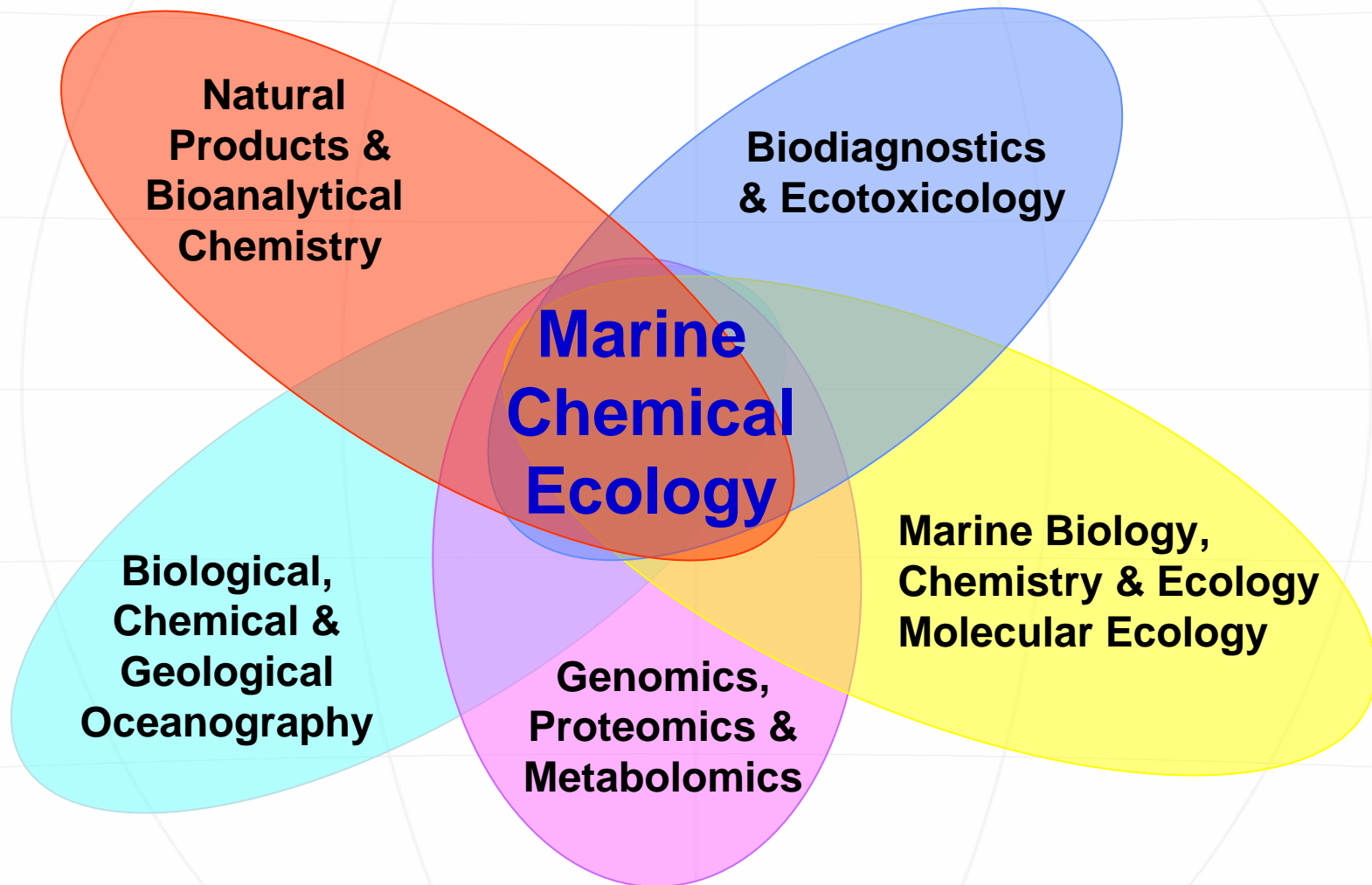
?

Defense against Predators,

Prymnesium parvum

Elimination of Competitors

Towards Inter-disciplinary Science



Strategies to answer these questions:

Can toxic strains be detected genetically?

=> Genomic characterization (microsatellites, AFLP, rDNA sequence analysis)

Which genes are responsible for growth and toxicity?

=> Gene expression analysis (EST, Data bases, microarrays)

What toxins are present?

=> Bioanalytics (LC-FD, LC-MS/MS)

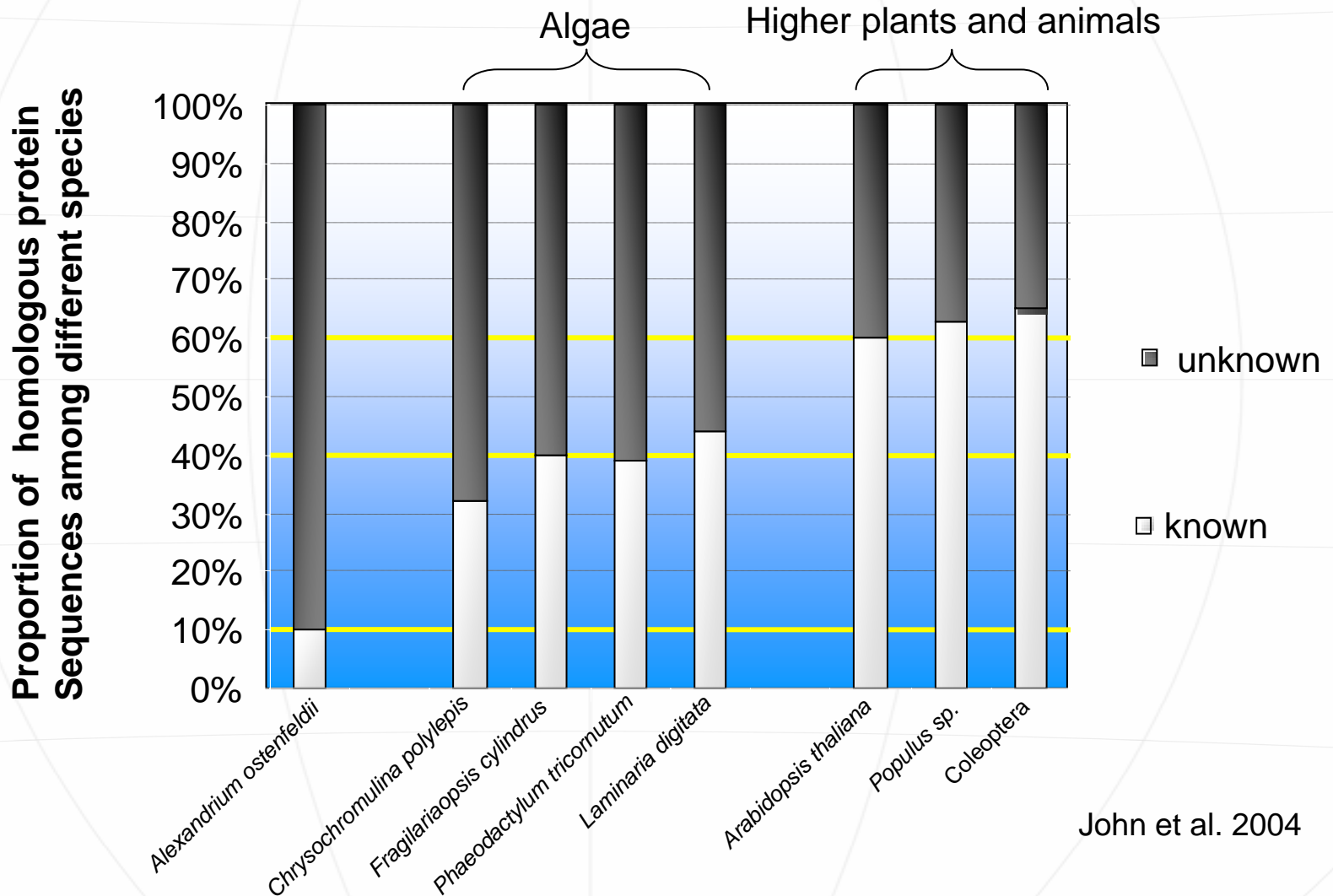
What are the allelochemicals?

=> Chemical experiments & bioassays

What effects do toxins have?

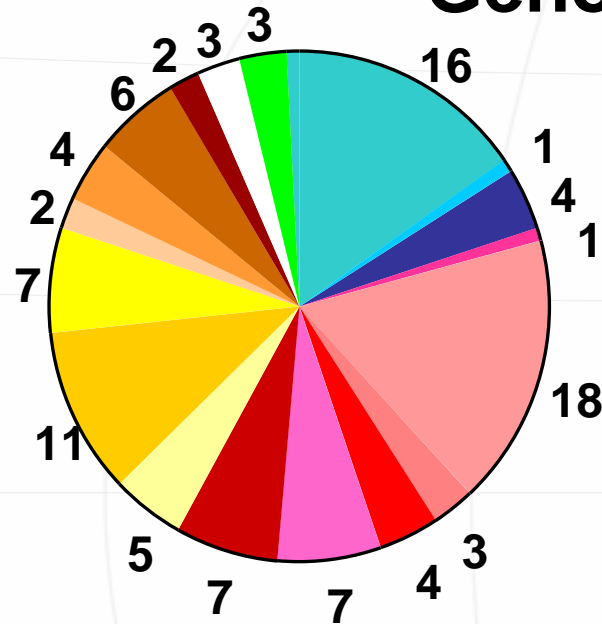
=> Toxicological assays

Genomic Characterization

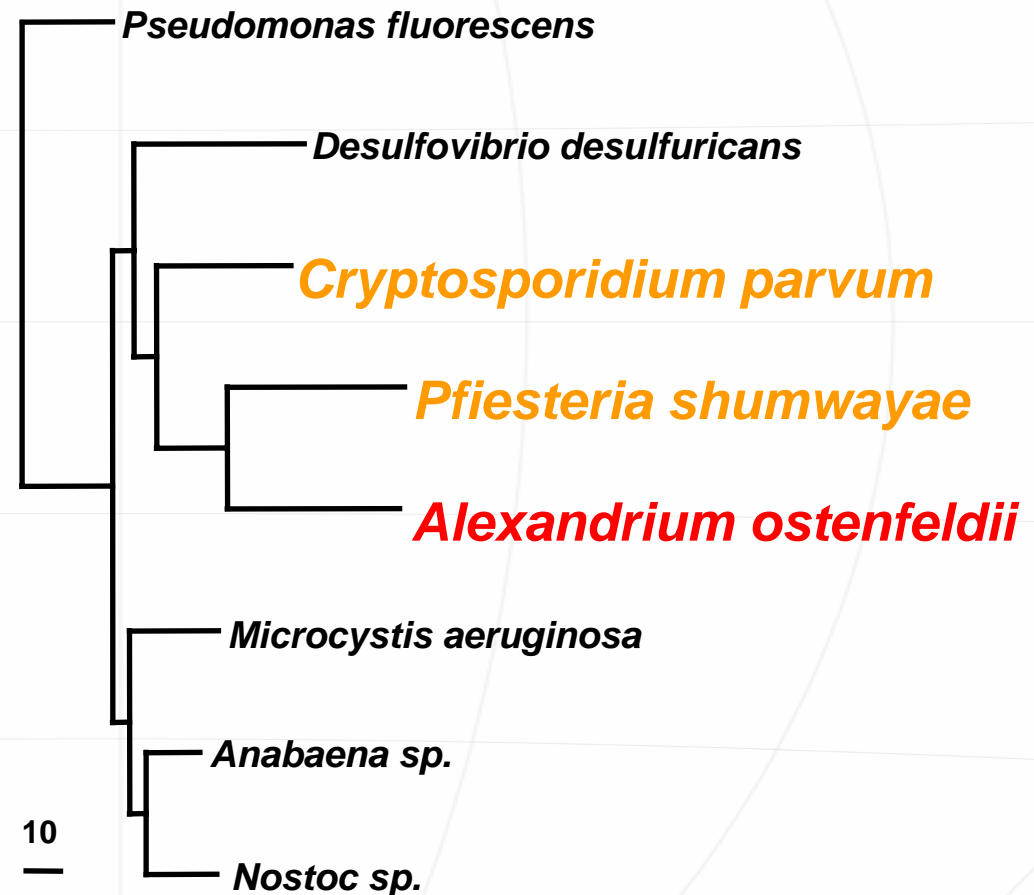


John et al. 2004

Genomic Characterization



PKS EST Analysis



Cell structure

Information storing and processing

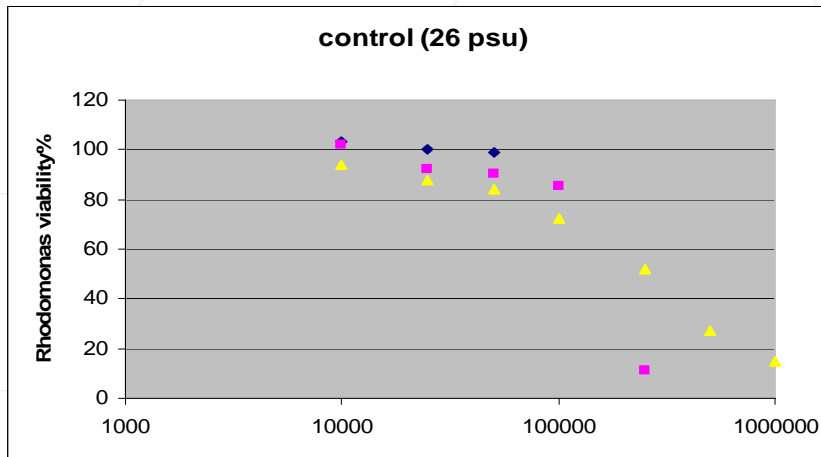
Cellular processes

Metabolism

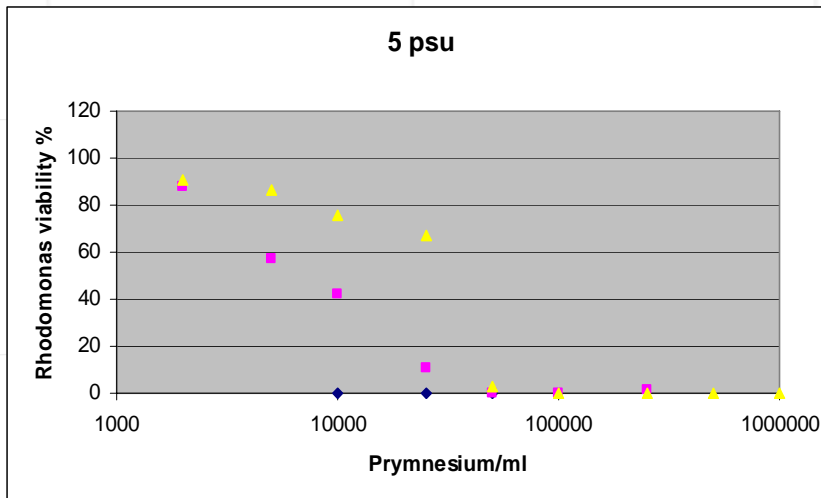
General function (prediction only)

Stress, defence and toxicity

Gene Expression Analysis



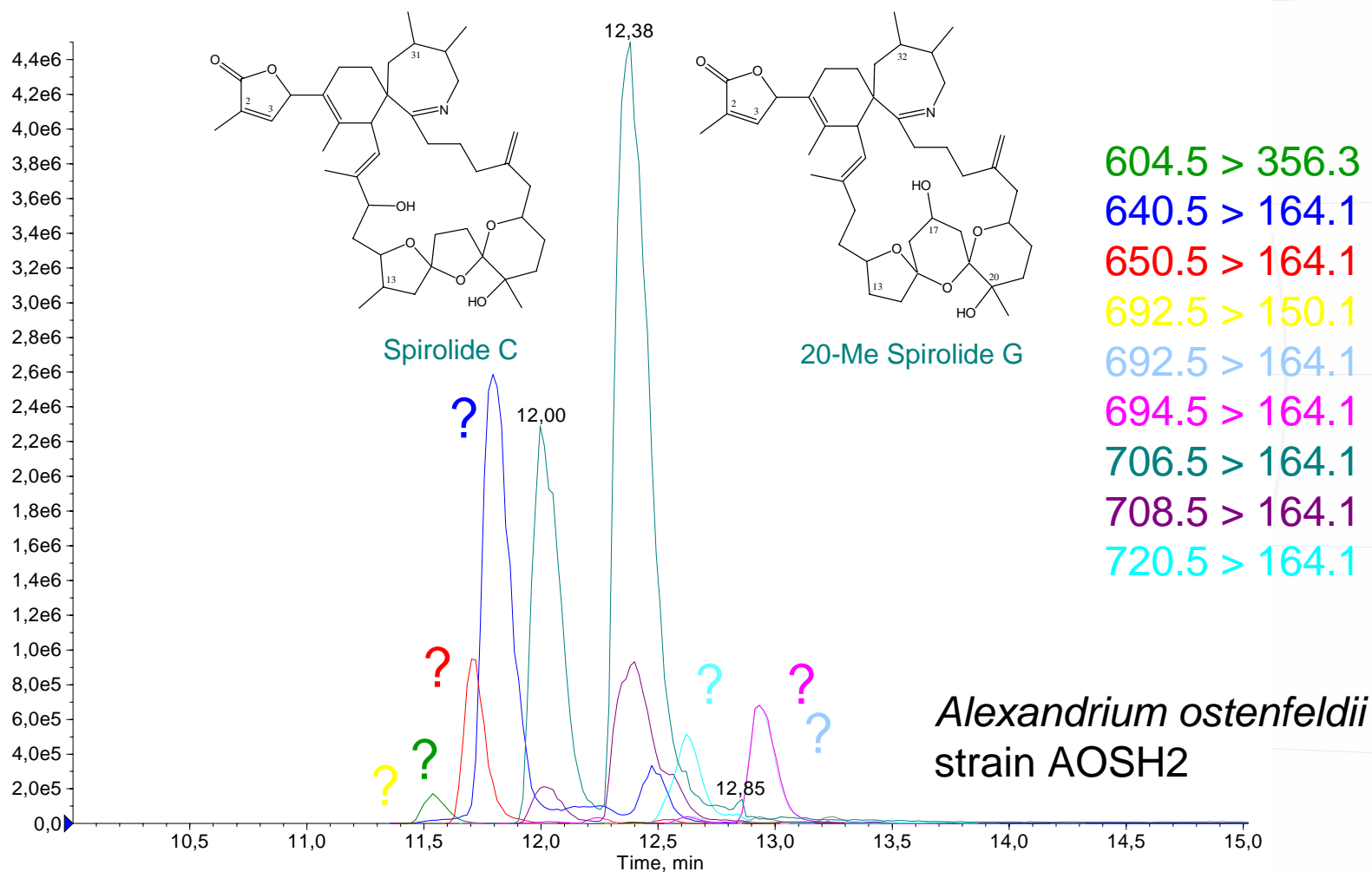
- Early exponential growth phase
- Late exponential growth phase
- Stationary phase



Toxicity of *Pymnesium parvum* is high at low salinity

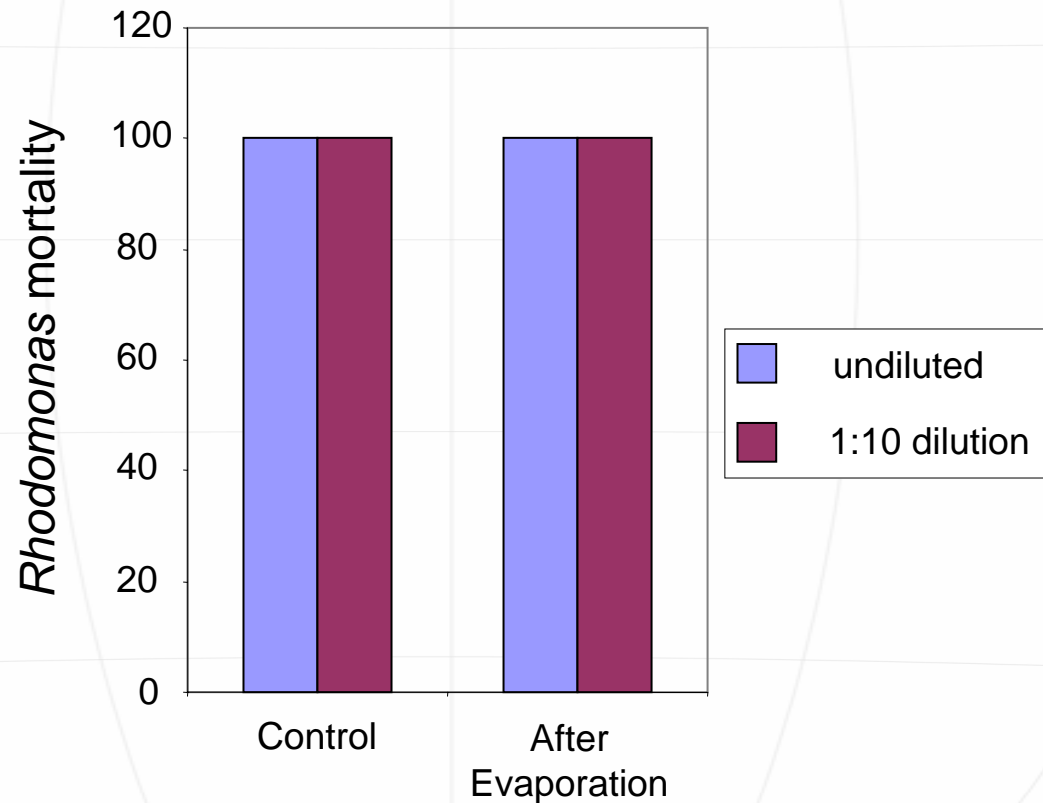
=> ESTs, Microarrays

Bioanalytics



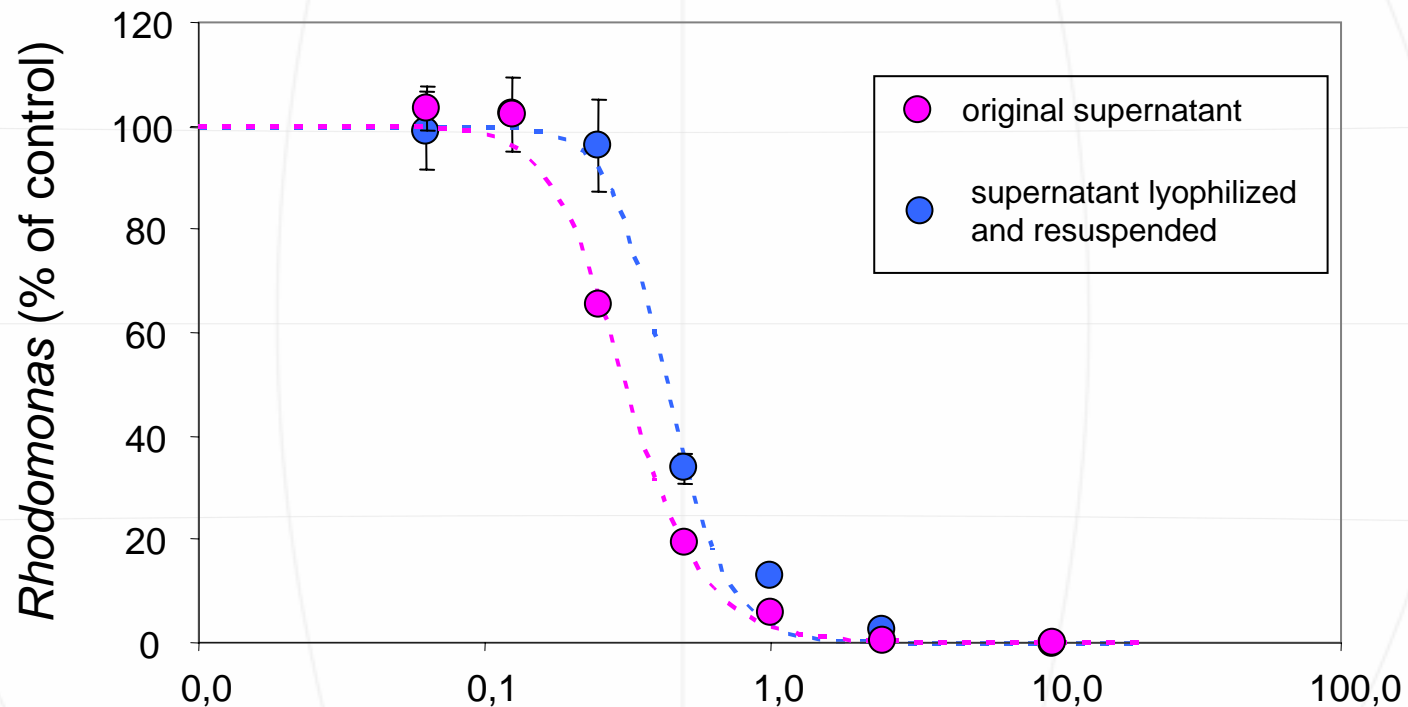
Chemical Experiments – Evaporation

Alexandrium tamarense supernatant – Lytic Effect on *Rhodomonas*



Chemical Experiments – Lyophilization

Alexandrium tamarense supernatant – Lytic Effect on *Rhodomonas*



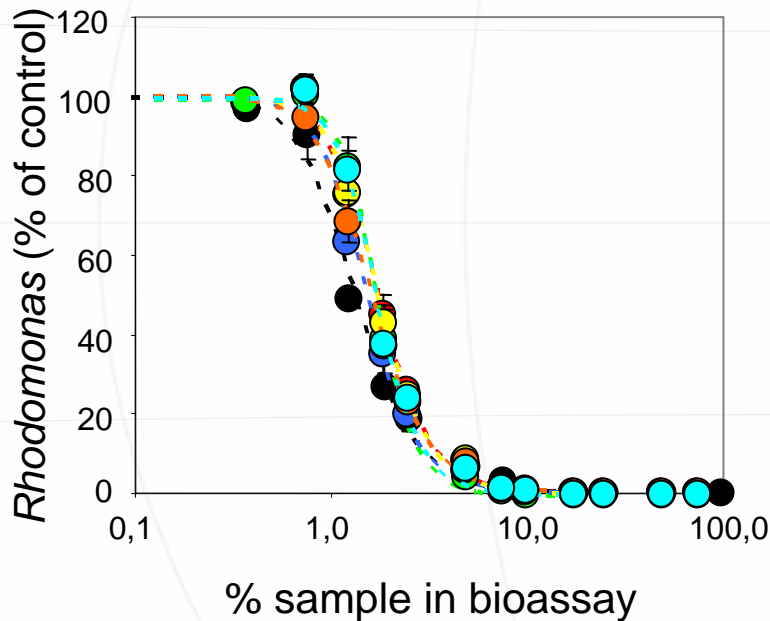
EC₅₀ original: 0.31 % % sample in bioassay

EC₅₀ „instant“: 0.44 %

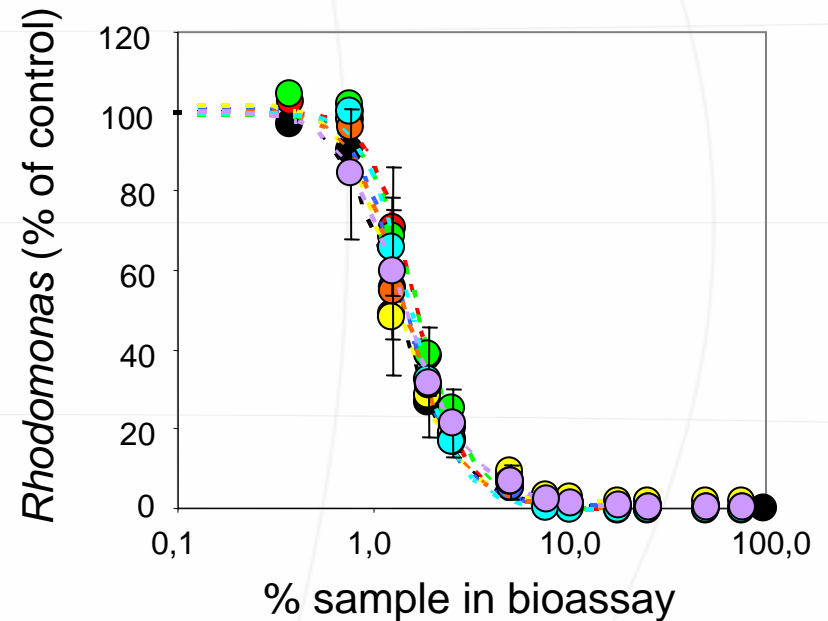
Chemical Experiments – Stability

Alexandrium tamarense supernatant – Lytic Effect on *Rhodomonas*

15°C; light (150 $\mu\text{E m}^{-2} \text{s}^{-1}$)



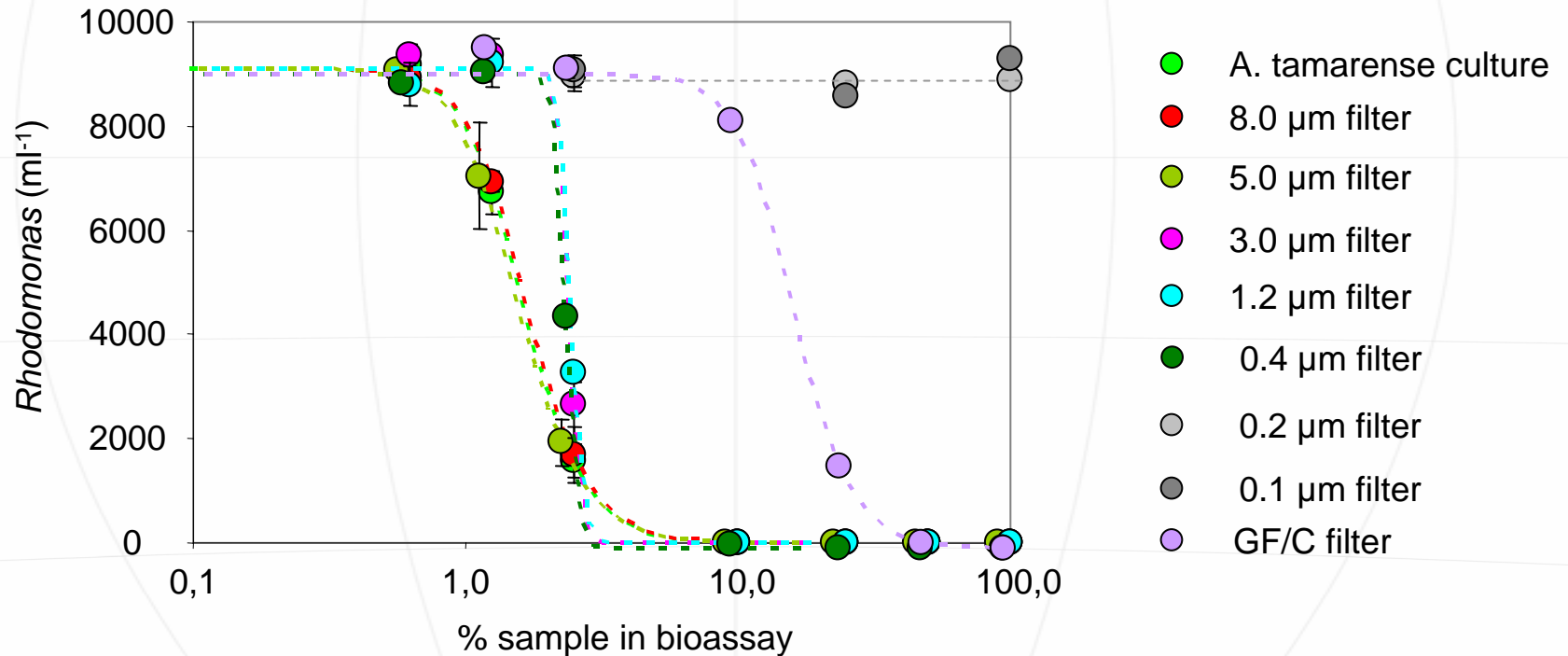
15°C; dark



- t = 0
- t = 4d
- t = 12d
- t = 49d
- t = 1d
- t = 7d
- t = 20d

Chemical Experiments – Filterability

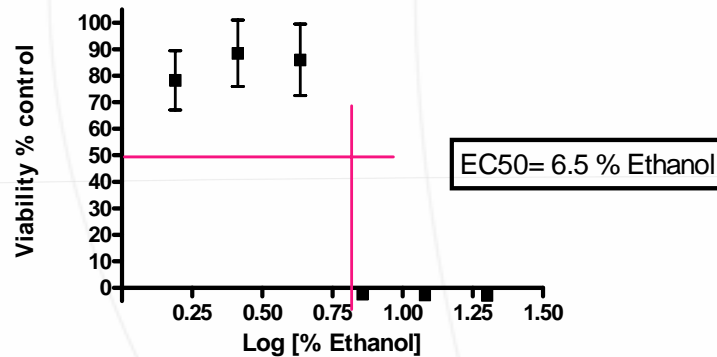
Alexandrium tamarensis supernatant – Lytic Effect on *Rhodomonas*



Toxicological assays

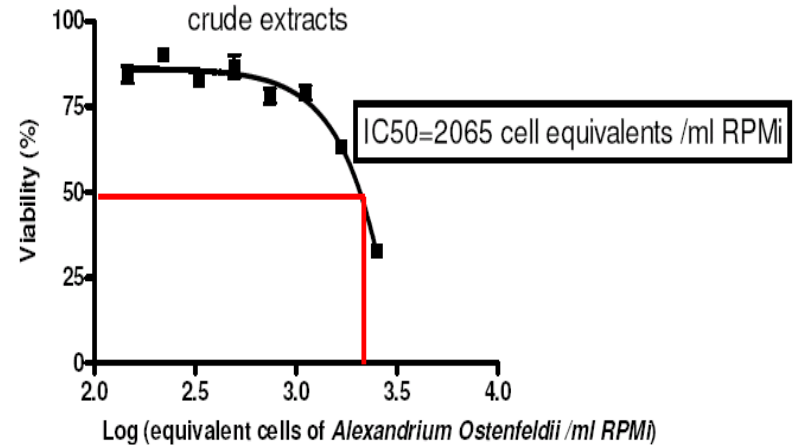
Alexandrium ostenfeldii extract – Toxic Effect on Neoblastoma cells

Viability curve of N2a exposed to Ethanol



Control

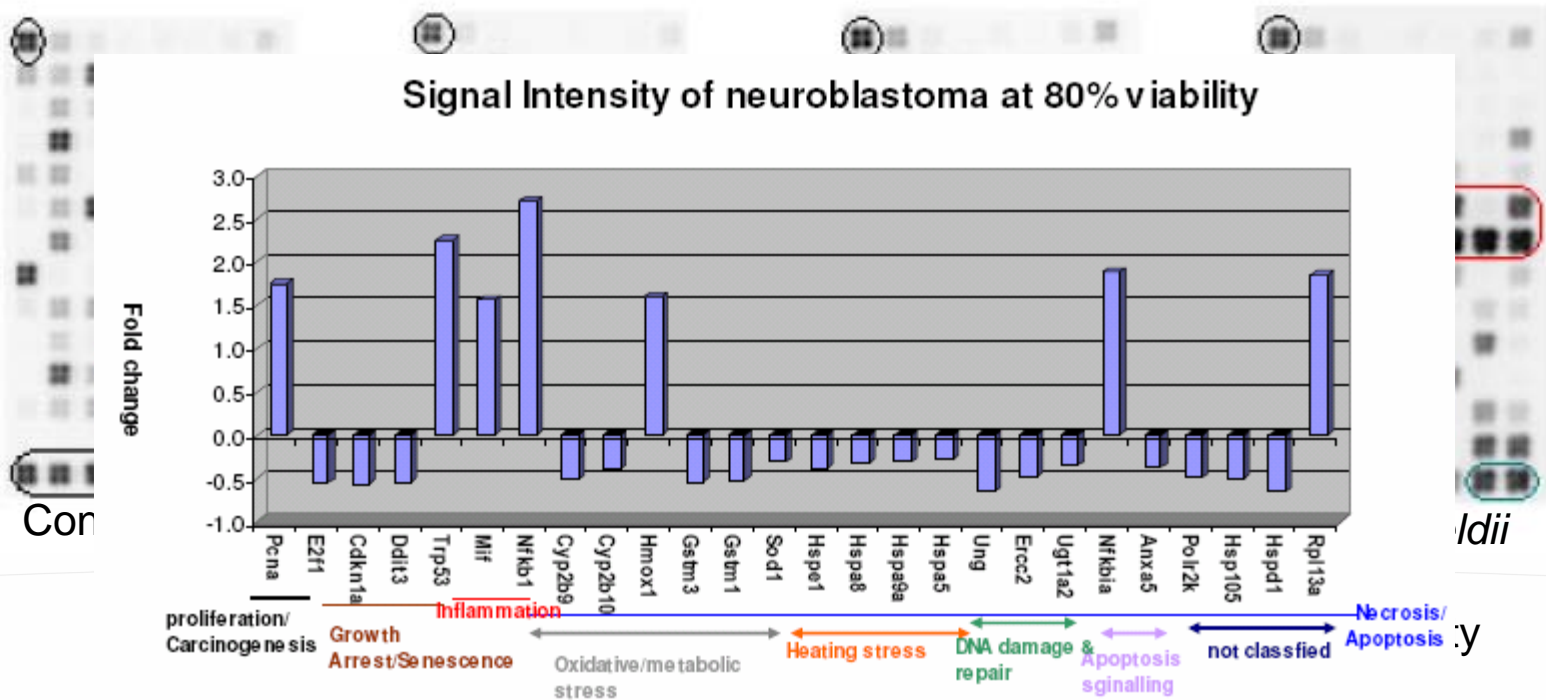
Viability curve of N2a exposed to *A. ostenfeldii* crude extracts



A. ostenfeldii cell extracts

Toxicological assays

Alexandrium ostenfeldii extract – Expression of stress and toxicity related genes (GEArray Q Series Mouse Stress & Toxicity Pathway Finder)



AWI

Allan D. Cembella
Uwe John
Urban Tillmann
Bernd Krock
Tilman Alpermann
Sascha Klöpfer
Ines Jung

Sára Beszteri
Nina Jaeckisch
Ines Marschallek
Chishimba M. Kantu
Chibo Chikwililwa
Annegret Müller
Wolfgang Drebing

GKSS

Andreas Prange
Jürgen Gandraß
Sandra Schäfer
Beritt Schwalger

Thank You
for
Your Attention!