# A Decision Support System for the ecological risk assessment of freshwater sediments and lotic systems

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# Current approach in environmental assessment by European legislations

Law limits for single chemicals

- Quantification of about **100-150** chemicals
- Comparison of concentrations with **safety limits**

However more than **280.000 substances** are registered as **toxic** by the American Chemical Society

Examples of **emerging pollutants**:

- pharmaceuticals
- body care products
- drugs
- etc…

# Limits of the chemical approach

- It's impossible to quantify **all potentially toxic substances**
- It's difficult to infer toxic effects due to **mixture of pollutants**
  (e.g. additive, synergistic, etc…)
- It's challenging to predict **bioavailability** of pollutants in different field conditions

# Coupling biological and chemical data

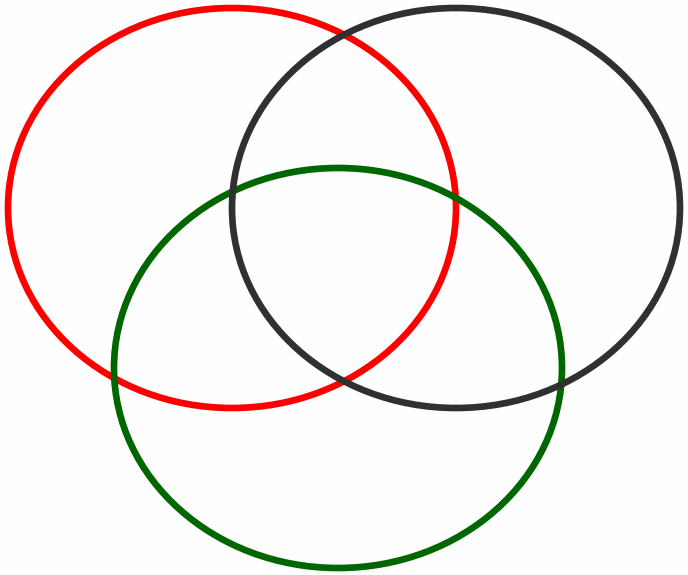
- Assessment of the **ecological risk** of a site

  à through a weight-of-evidence **Triad approach** (i.e. integration of chemical, ecotoxicological and ecological data)
- Evaluation of the **quality** of an environmental matrix, such as sediment and water

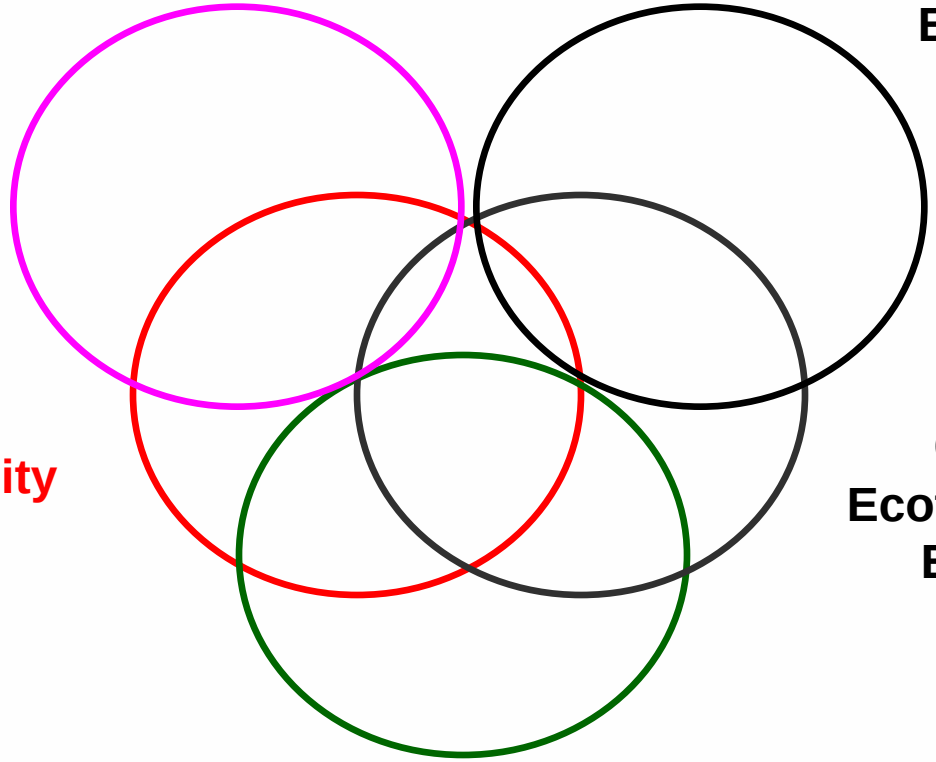
  à through a 2-legs **Duade approach** (i.e. integration of chemical and ecotoxicological data)

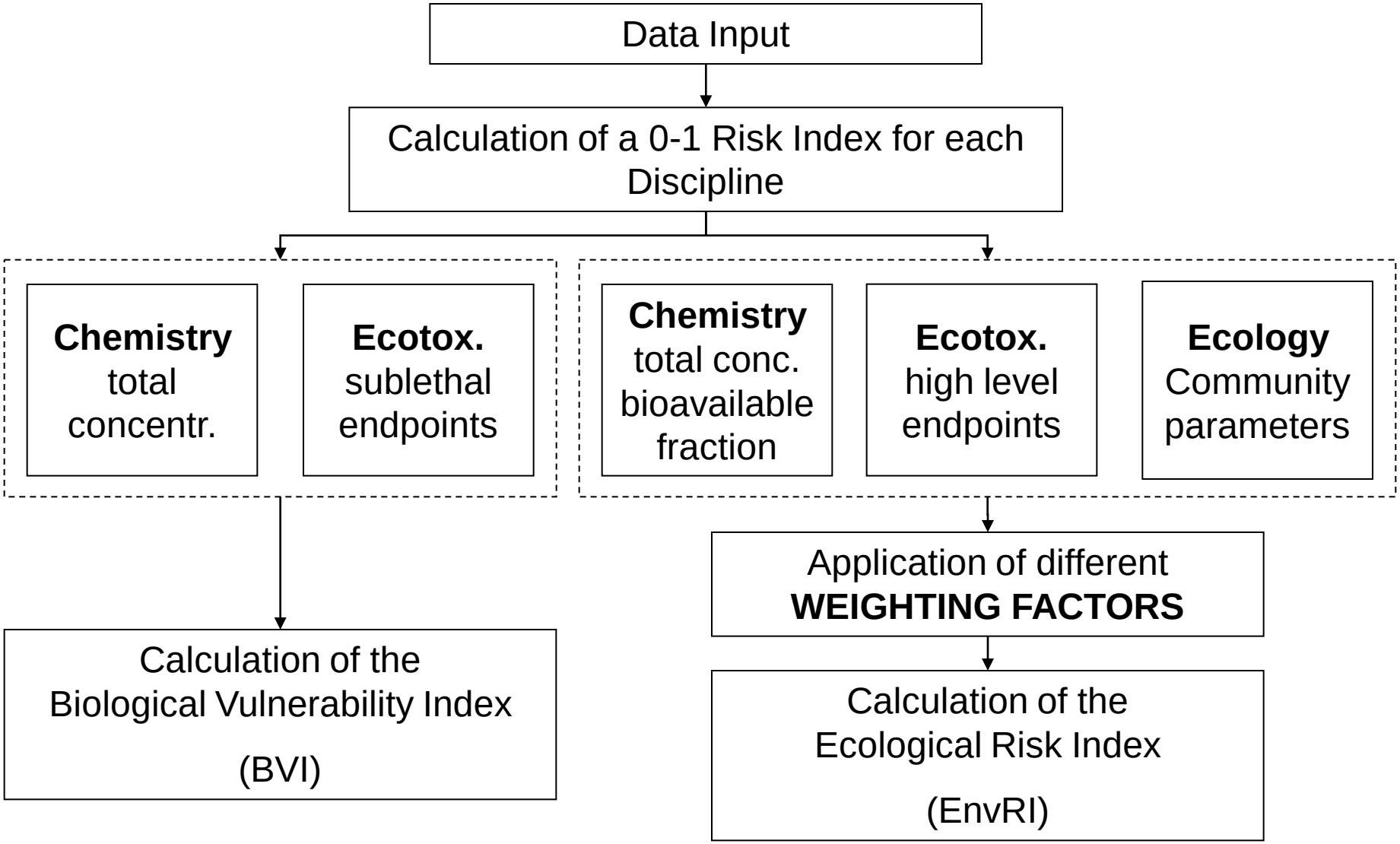
Crucial for a **correct environmental management** (e.g. dredging activities of dams, remediation, etc…)

# The Triad approach

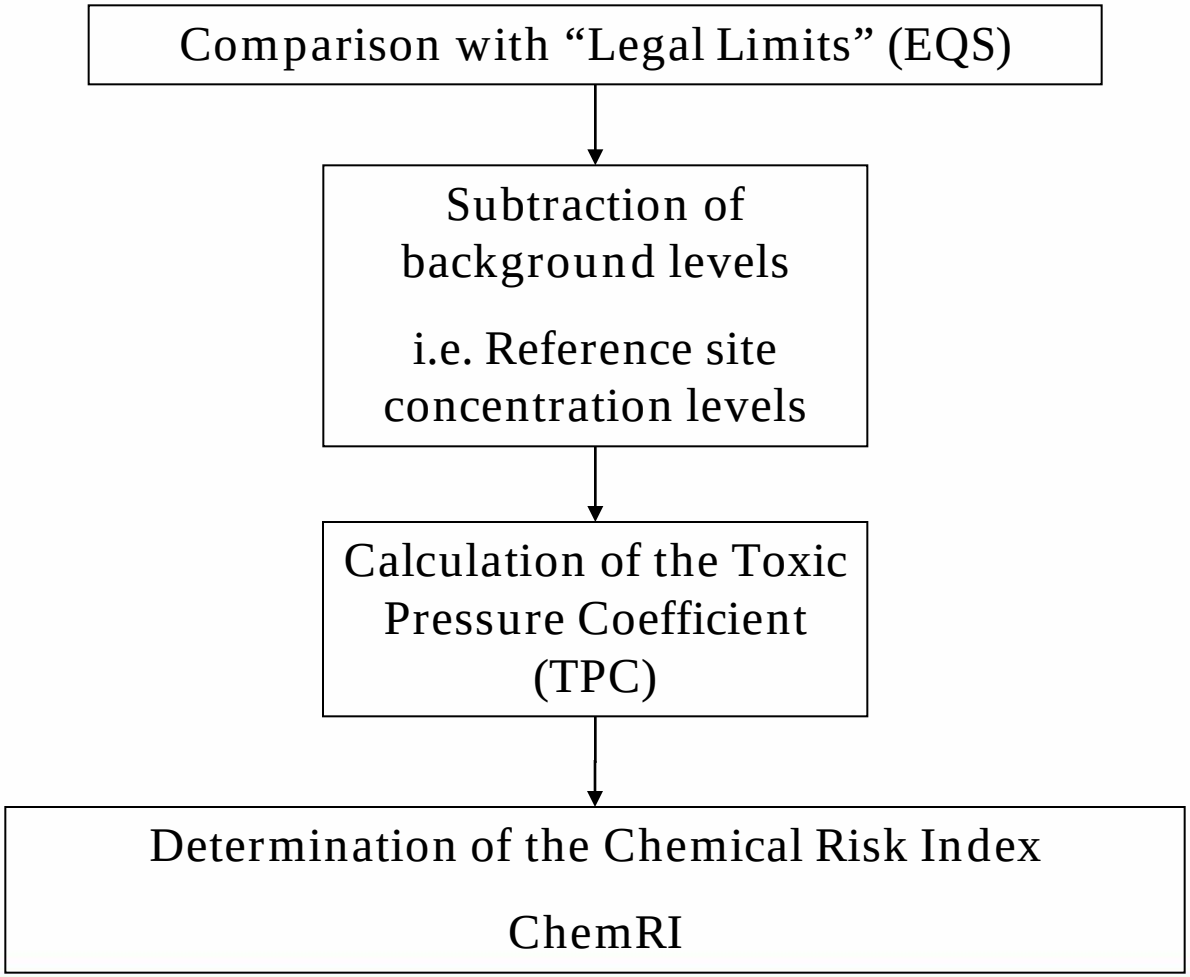


# The Triad approach

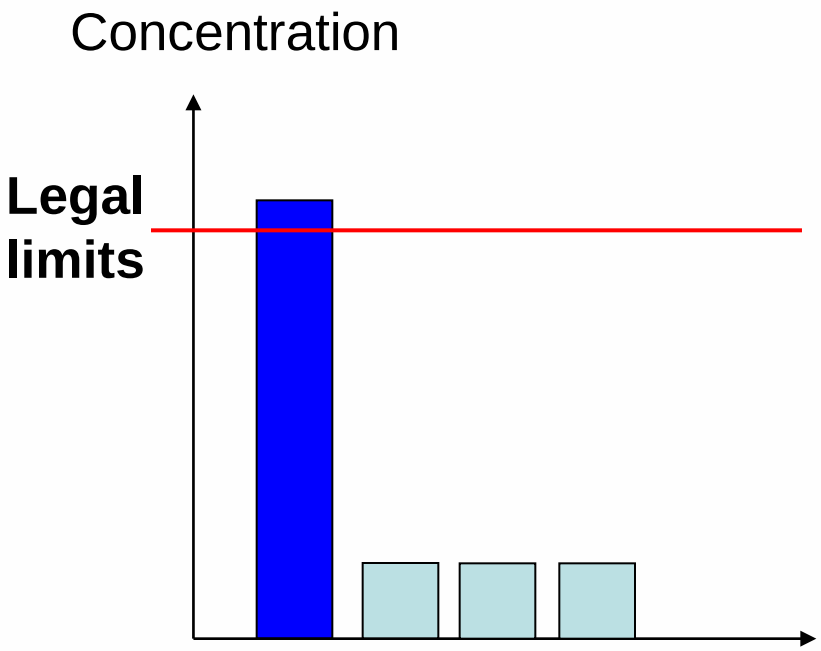


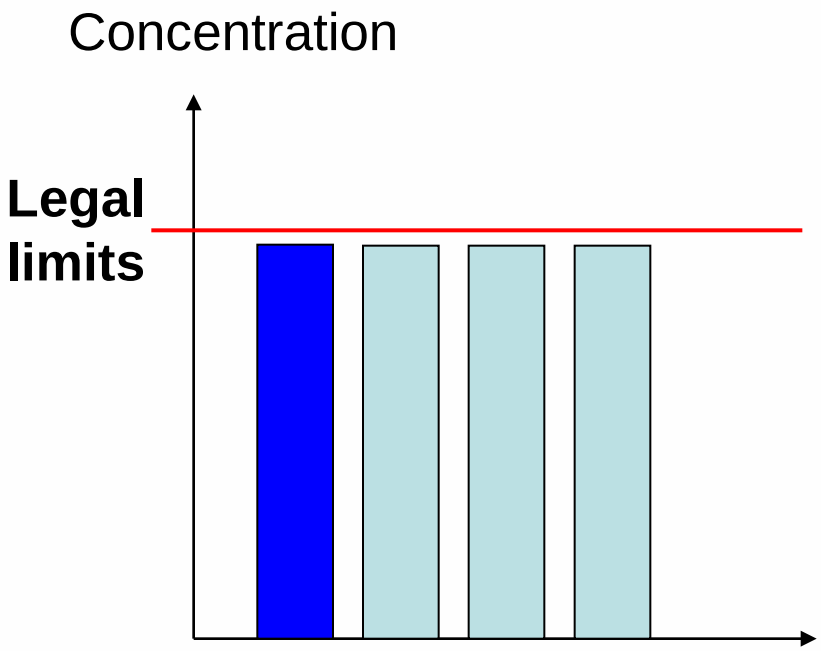

Data Input
Calculation of a 0-1 Risk Index for each Discipline
Chemistry total concentr.
Ecotox. sublethal endpoints
Chemistry total conc. bioavailable fraction
Ecotox. high level endpoints
Ecology Community parameters
Application of different WEIGHTING FACTORS
Calculation of the Biological Vulnerability Index (BVI)
Calculation of the Ecological Risk Index (EnvRI)

# Chemical Risk Index



# Limits of “single compound” thresholds

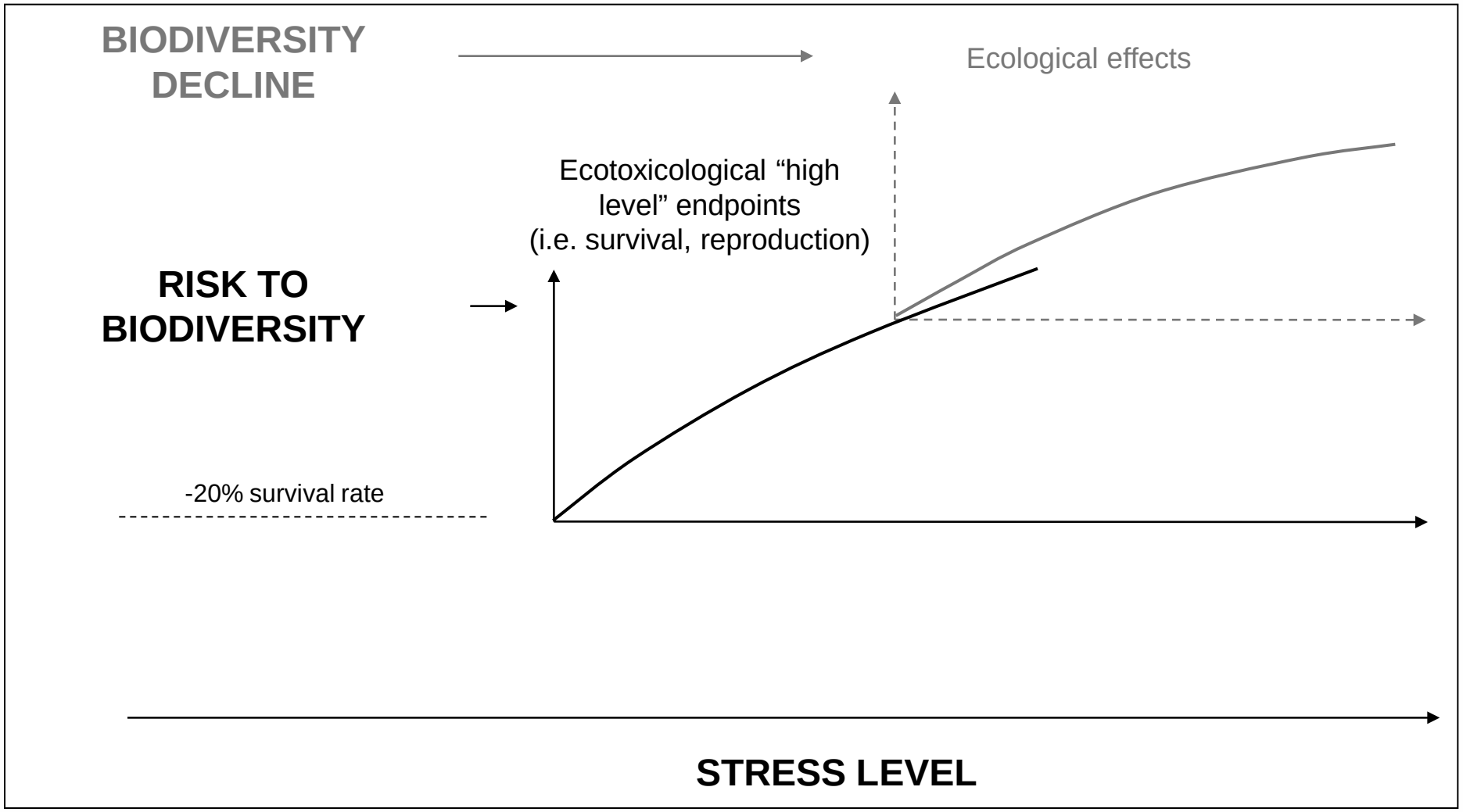


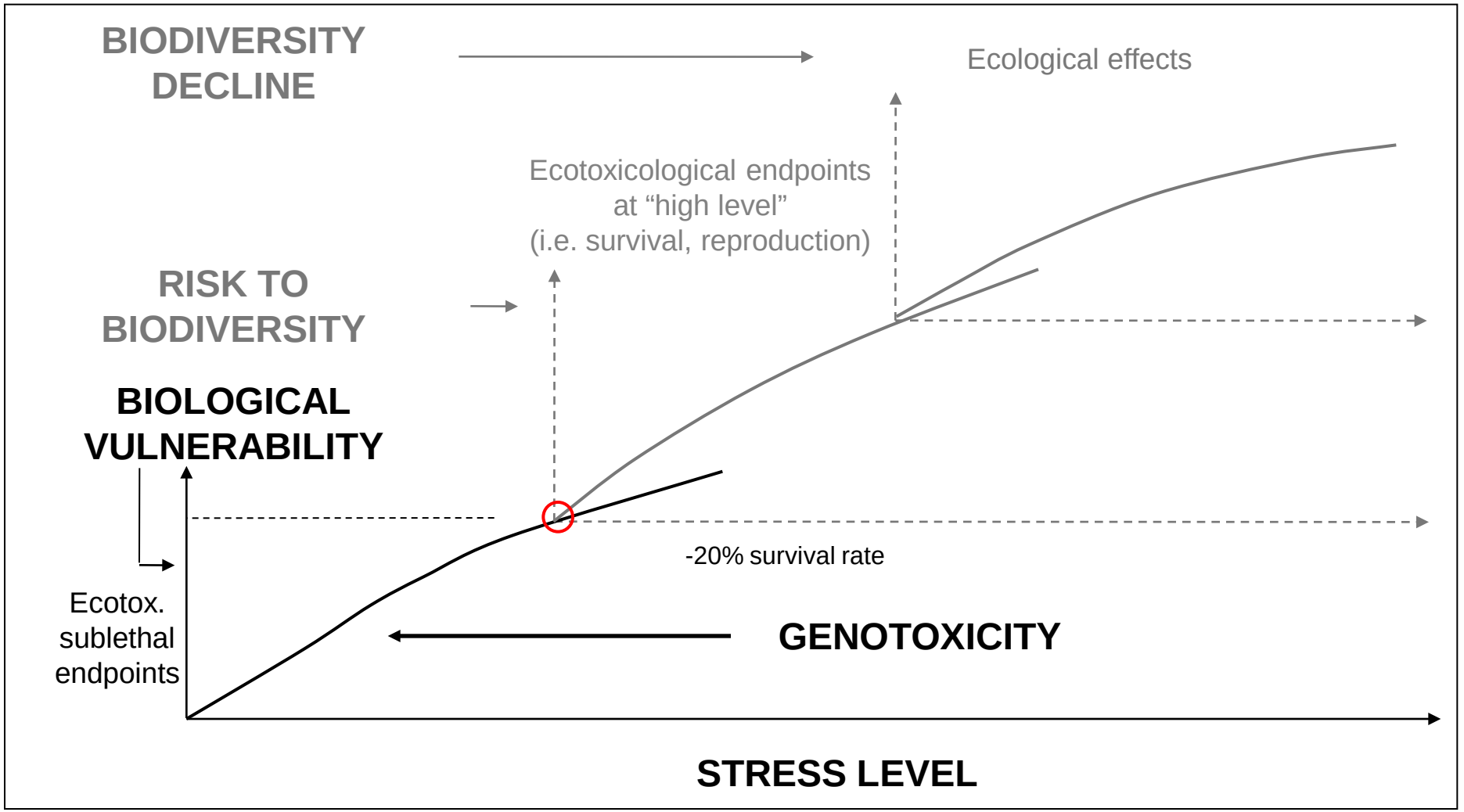



**BIODIVERSITY DECLINE**

Ecological effects

**STRESS LEVEL**


BIODIVERSITY DECLINE
Ecological effects
Ecotoxicological “high level” endpoints (i.e. survival, reproduction)
RISK TO BIODIVERSITY
-20% survival rate
STRESS LEVEL

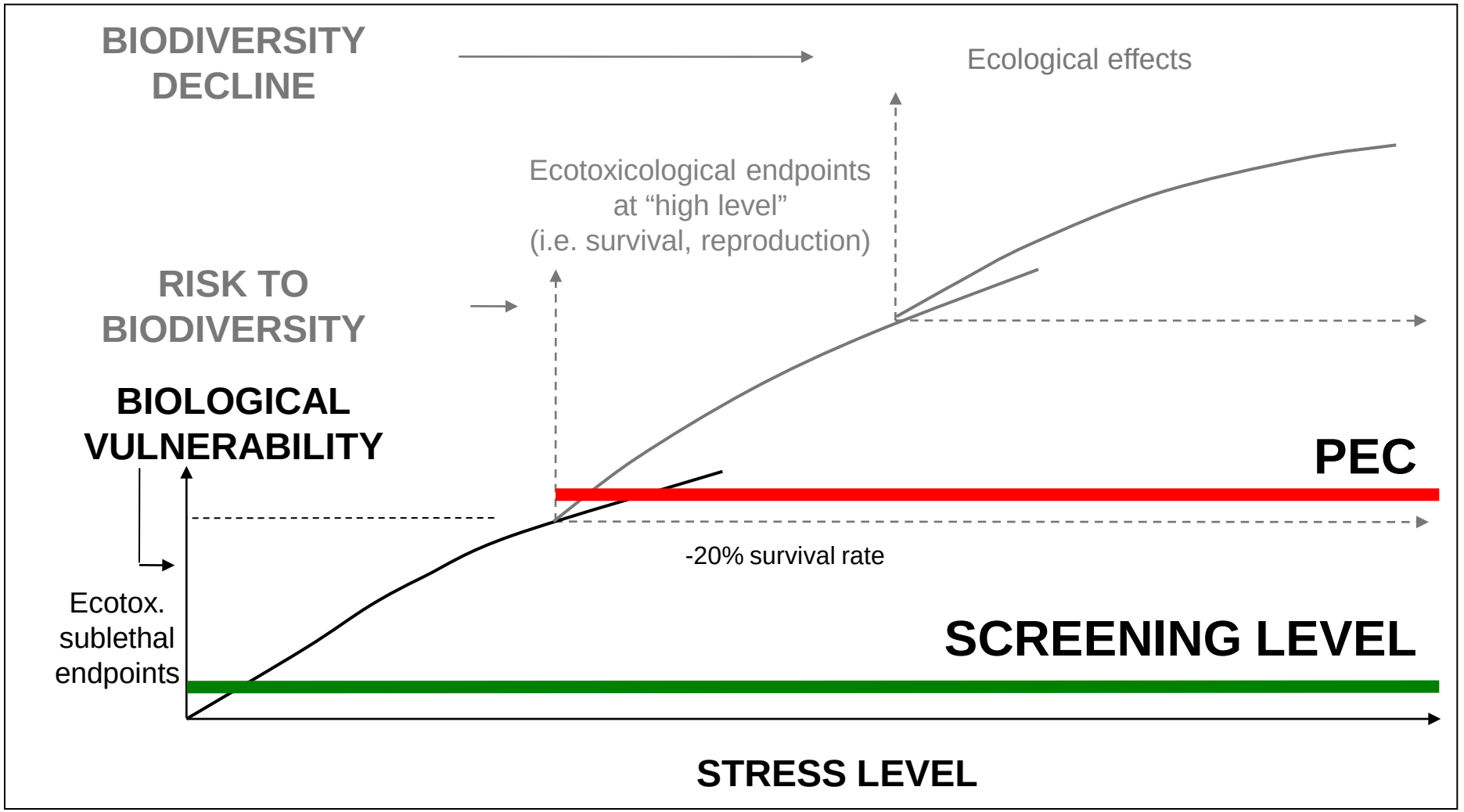

BIODIVERSITY DECLINE
Ecological effects
Ecotoxicological endpoints at "high level" (i.e. survival, reproduction)
RISK TO BIODIVERSITY
BIOLOGICAL VULNERABILITY
-20% survival rate
Ecotox. sublethal endpoints
GENOTOXICITY
STRESS LEVEL

## Chemical approach: comparison with two threshold values

- Screening level
  No effect concentration
  (octanol/water ratio; McDonald et al, 2000)
- PEC (Probable effect concentration)

## Risk-based approach: use of Duade / Triad tools

- **Dredged sediment**
  Duade – 2-tiers approach
  (chemical and ecotoxicological data)
- **River environments**
  Triad – 2-tiers approach
  (chemical, ecotoxicological and ecological data)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RISK TO BIODIVERSITY
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PEC
-20% survival rate
Ecotox. sublethal endpoints
SCREENING LEVEL
STRESS LEVEL

# Aquatic ecosystem

**Benthic macroinvertebrates**

*Extended Biotic Index* (different microhabitat)

**Microbial community**

*Bacterial biomass*, *DGGE*

**Diatom community**

*EPI-D*

# Aquatic ecosystem

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*EPI-D*

# Terrestrial - Riparian ecosystem

**Soil microarthropods**

*QBS index*

**Nematode community**

*Maturity index*

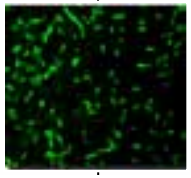
**Microbial community**

*Bacterial biomass*, *DGGE*

## WATER AND SEDIMENT

## SOIL

*Vibrio fischeri*



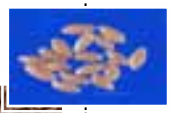
*Caenorhabditis elegans*

*Chiromons tentans*

*Eisenia andrei*

*Dictyostelium discoideum*

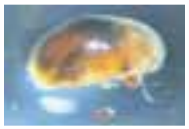
*Enchytraeus crypticus*



Phytotest

*Daphnia magna Straus*

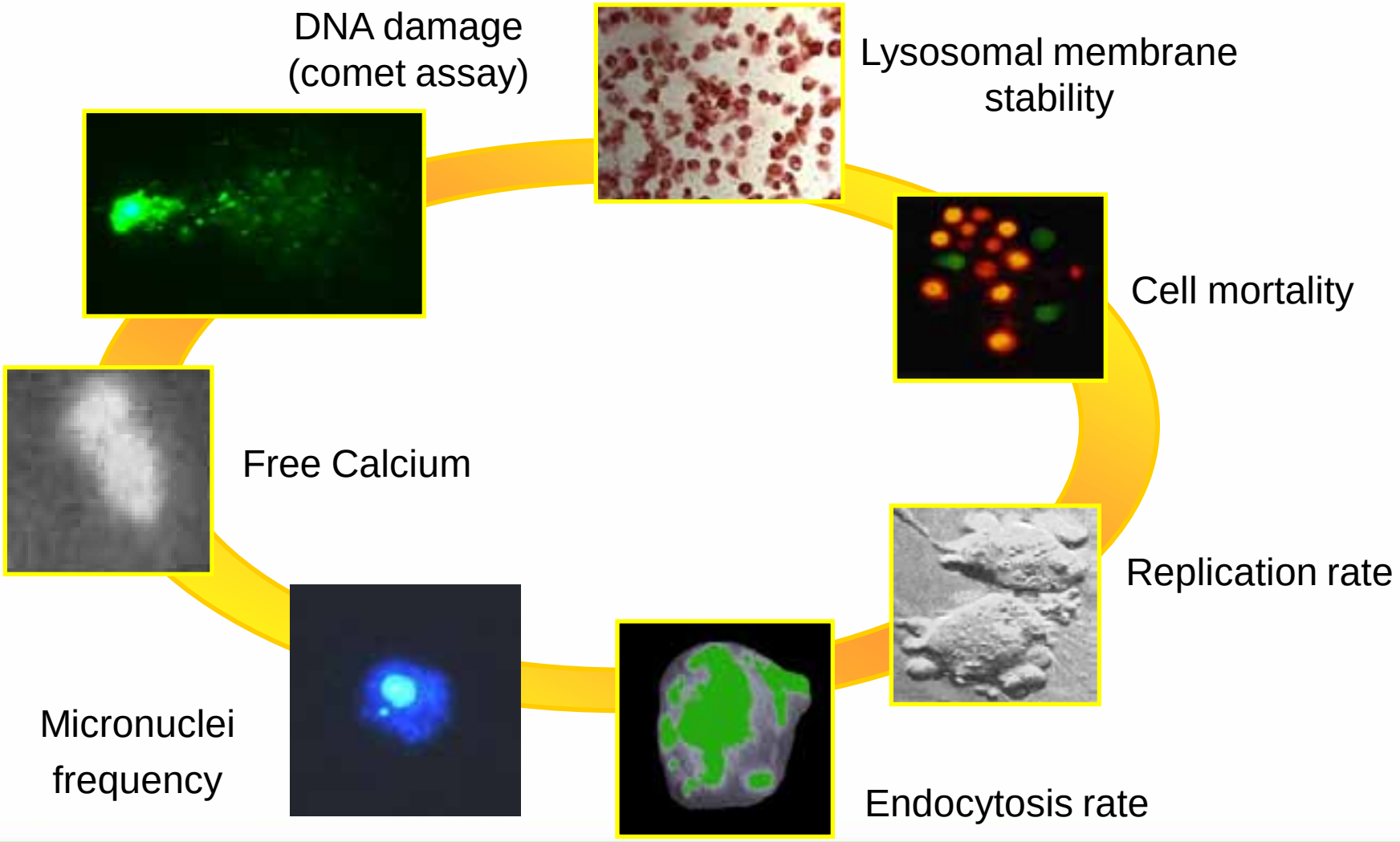
*Heterocypris incongruens*



*Folsomia candida*

*Pseudokirchneriella subcapitata*

# Assessment of sublethal effects
# Test with Dictyostelium discoideum



# Assessment of sublethal effects
# Test with Pisum sativum

Germination rate

DNA damage

(Comet assay)

Root growth

Mitotic index

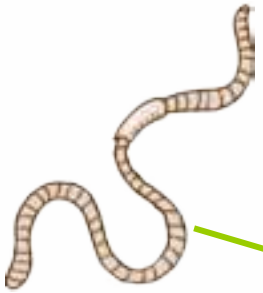
Micronuclei frequency

Mitotic anomalies

# Assessment of sublethal effects
## Test with earthworms

*Eisenia andrei*

*Lumbricus rubellus*



**Histology**

**Cytochemistry**



**Earthworm's digestive system**



**Plasmembrane CaATPase activity:**
**A – chloragogen tissue, B – intestinal epithelium**

**Stress biomarkers**

- **Lysosomal membrane stability**
- **Lipofuscine and neutral lipids accumulation**
- **$Ca^{2+}$-ATPase activity**
- **Lisosome/cytoplasm – Tissue damage**

**Exposure biomarkers**

- **MT: heavy metals response**
- **Peroxisomes proliferation**

**Genotoxicity biomarkers**

- **DNA damage (Comet assay)**
- **Micronuclei frequency**

Assessment of the **health status**

applying an

Expert System

# Molecular Highthroughput Techniques and The Systems Toxicology Approach

i.e. an integration of

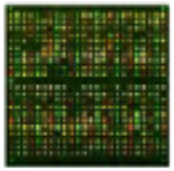
Transcriptomics

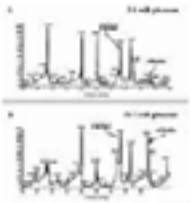
Proteomics

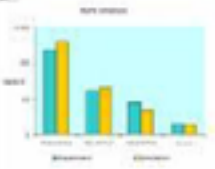
Metabolomics

biochemical / cytochemical /

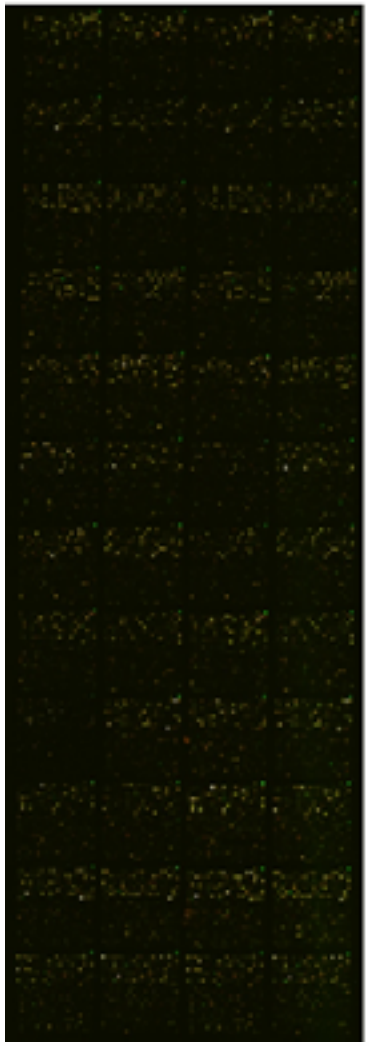
functional data (physiomics)







To explain mechanistic effects of pollutants in ecotoxicological relevant species



# What's before mortality?

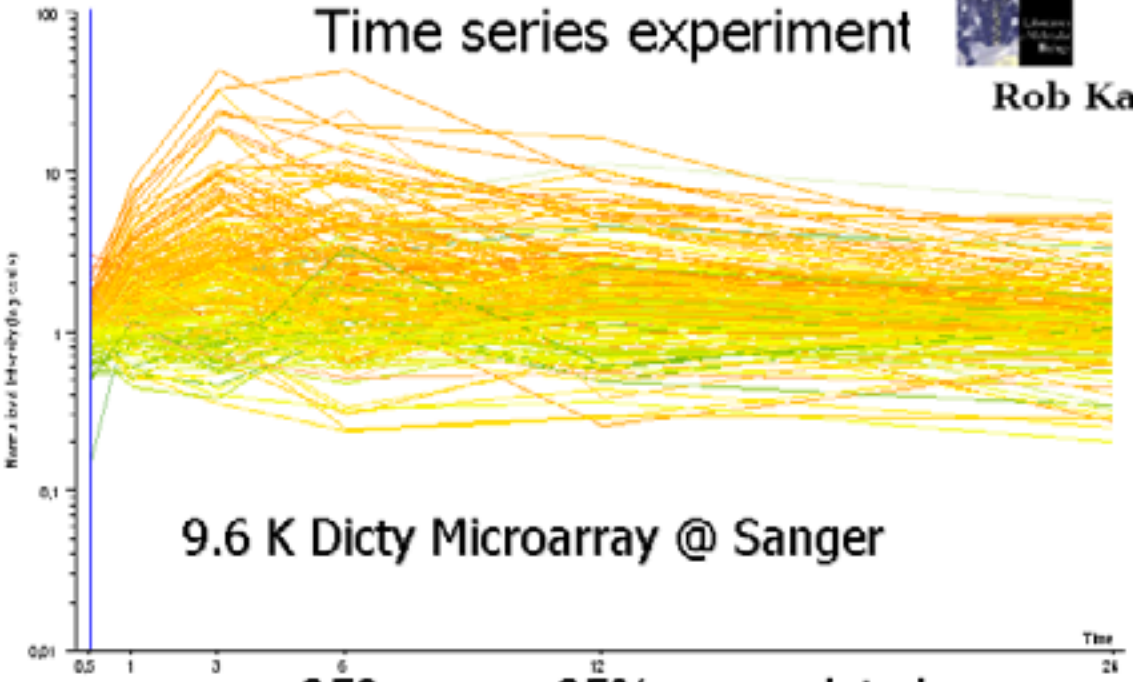
## Transcriptomic changes

The Wellcome Trust Sanger Institute

Al Ivens
Gareth Bloomfield
Jason Skelton

Rob Kay

Time series experiment




9.6 K Dicty Microarray @ Sanger

~ 250 genes; 85% upregulated

Homeostatic compensation

# Proteomics approach

Lysis and separation on 2DE gel (mainly cytosolic proteins)

↓

Digestion of the spots

↓

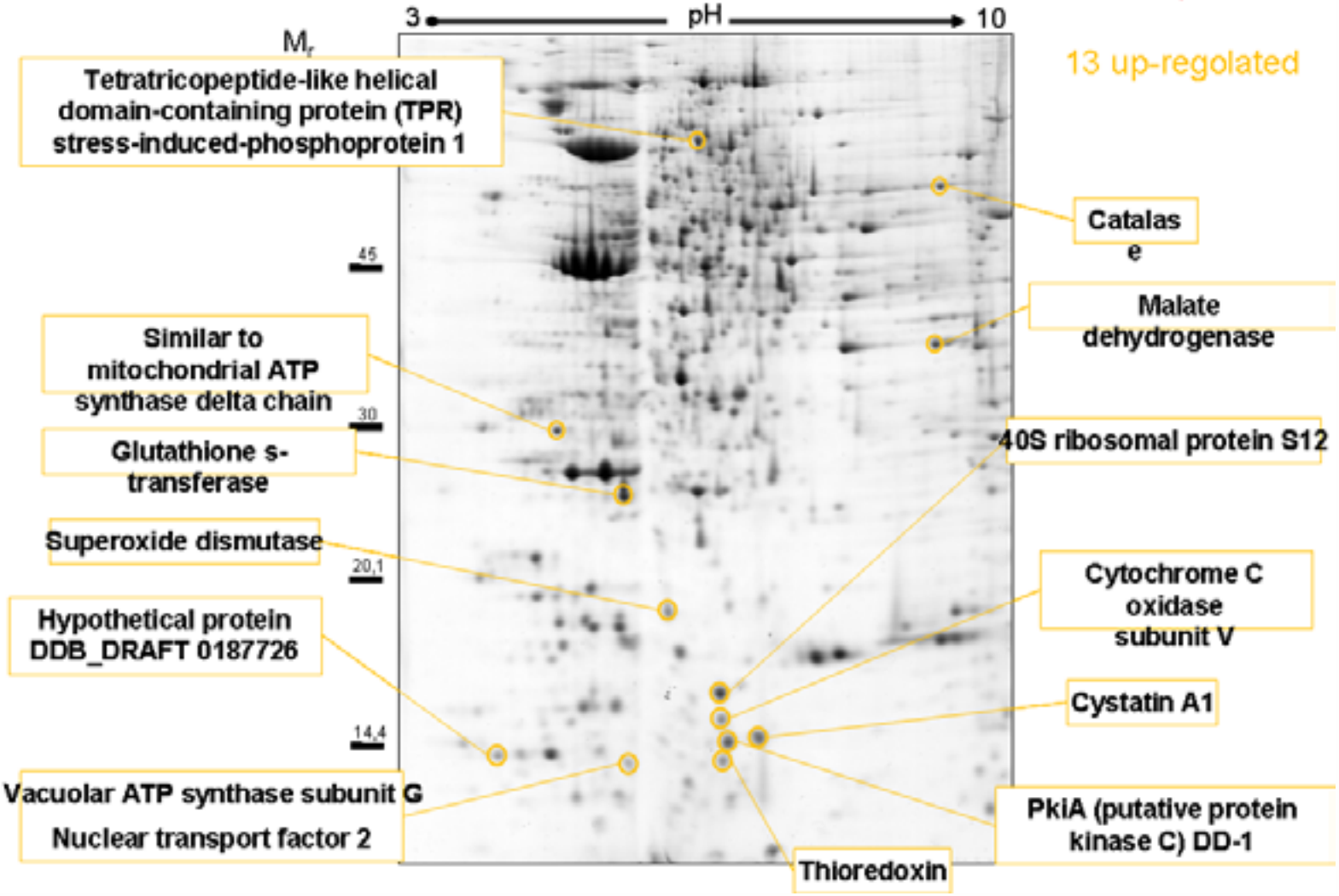
Identification by means of ESI-Q-TOF MS/MS

# 300 proteins were identified

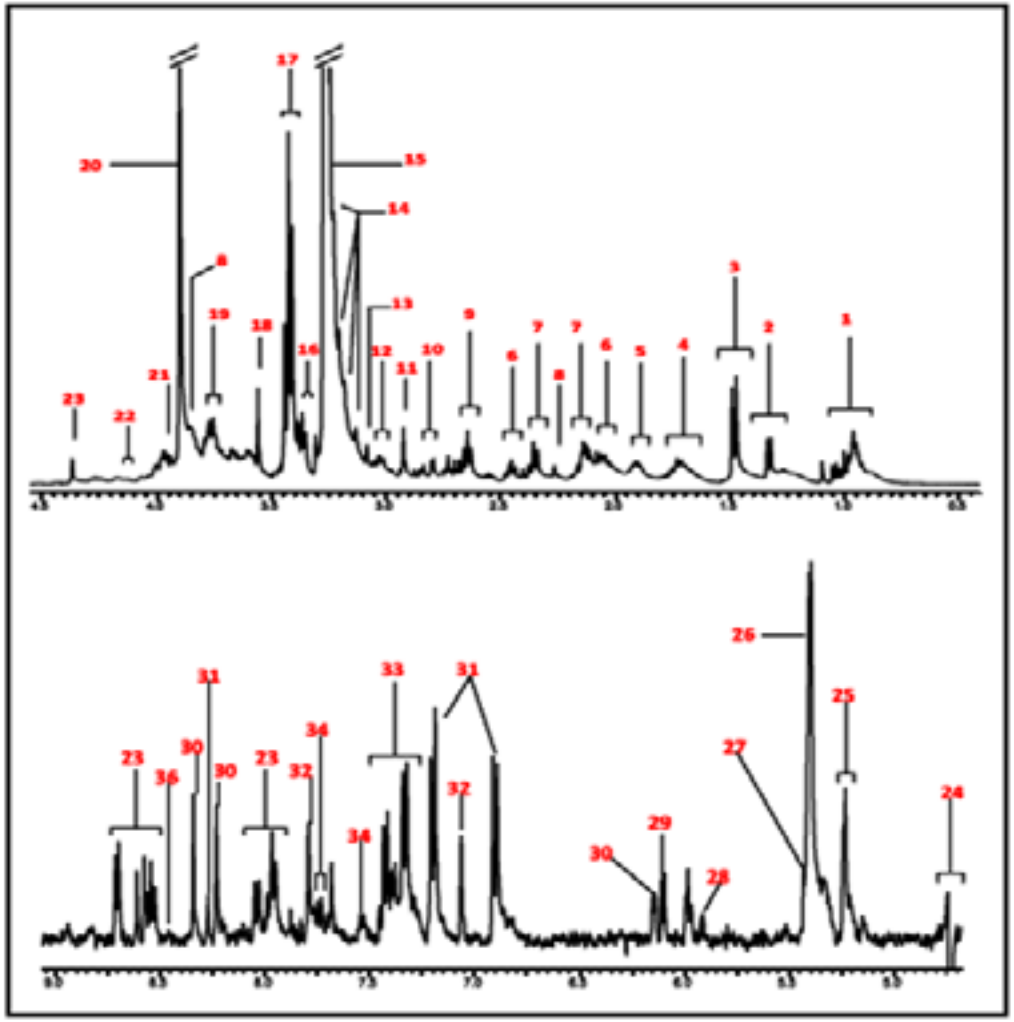
*Dictyostelium discoideum* – *Hg 2 µM*

13 spots:

13 up-regolated



# NMR spectra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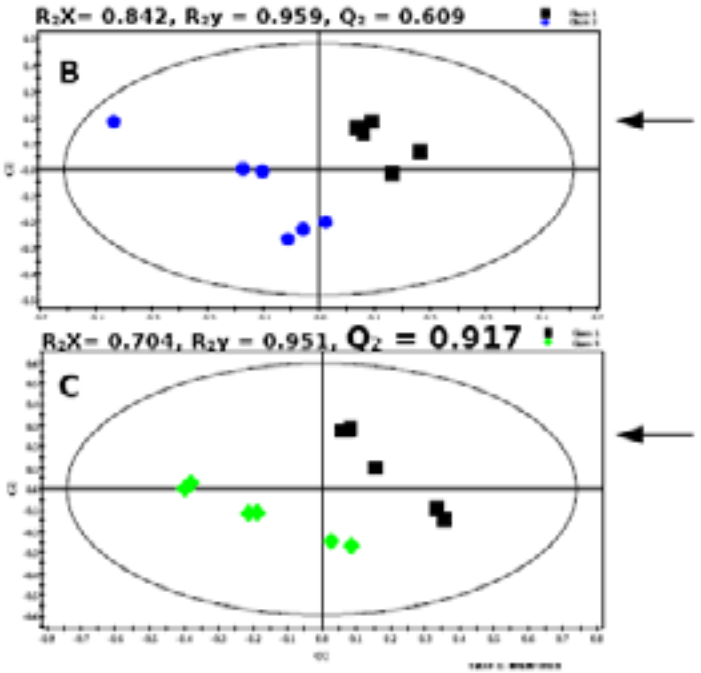
# Metabolomics

UNIVERSITY OF CAMBRIDGE
Jules Griffi
Oliver Jones

## Distinct patterns have been evidenced




Driving metabolite: increased levels of reduced GSH

Driving metabolites: drop of many aminoacids (serine/ glycine; valine, leucine, asparagine

# The systems toxicology approach suggested: