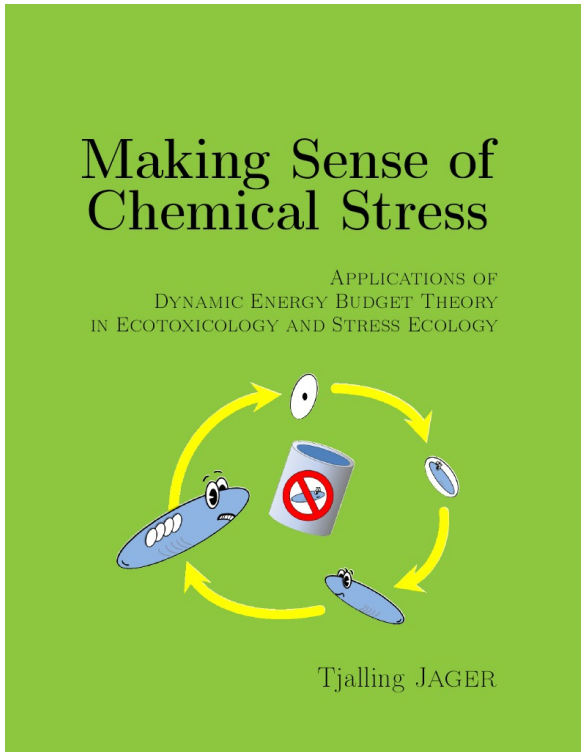
TKTD modelling
- Theoretical aspects –



Inspired from Jager (2015)
Making sense of chemical stress: application of dynamic energy budget theory in ecotoxicology and stress ecology.

Leanpub: https://leanpub.com/debtox_book, Version 1.2.

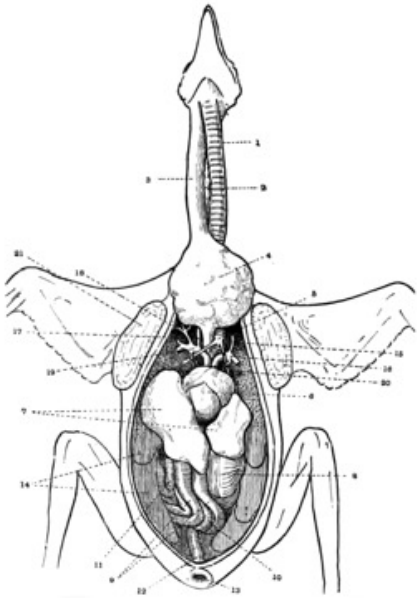
https://www.ecotoxmodels.org/hot-topics/toxicokinetic-toxicodynamic-models/

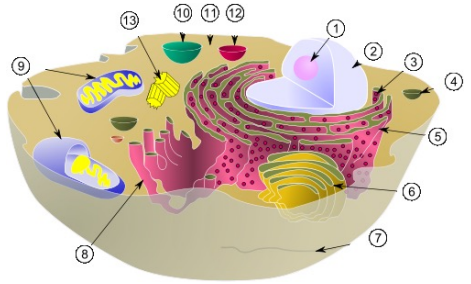
What means TKTD?

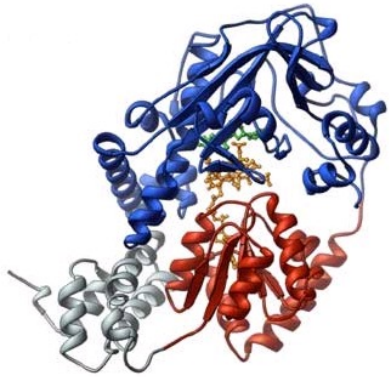
- **TK stands for Toxico-Kinetics**
  - Provides a prediction of the chemical concentration inside an organism from the external chemical concentration to which the organism is exposed;
  - Models absorption, distribution, metabolism (biotransformation) and elimination of toxicants inside the organism (ADME processes);
  - Includes physiologically-based (PB) processes.

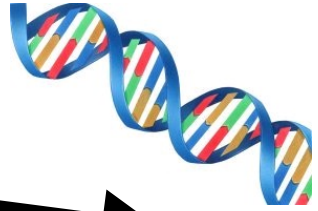
- **TD stands for Toxico-Dynamics**
  - Translates the internal chemical concentration to an effect on life-history traits over time (e.g., survival, growth, reproduction,…);
  - Accounts for energy allocation and physiological compensation;
  - Accounts for damage inside the organism.

Organisms are complex …









Stressing organisms…

… adds to this complexity

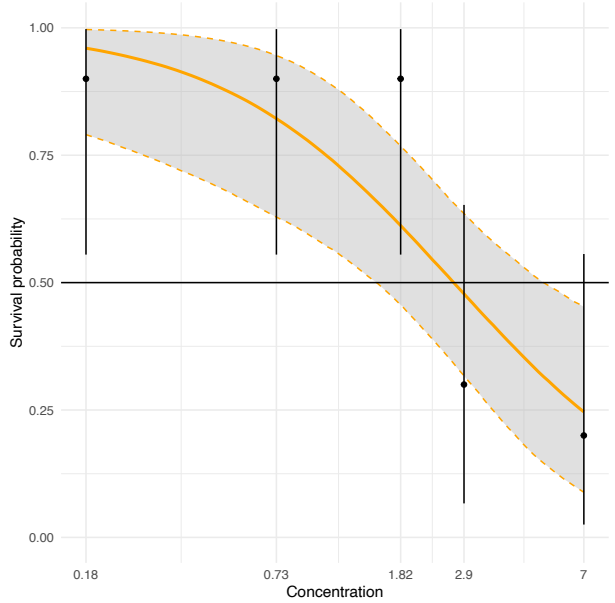
Response to a chemical stressor depends on:

- type of toxicant;
- organism (species, life stage…);
- endpoint (survival, growth, reproduction…);
- exposure duration and intensity;
- environmental conditions.

How to face with this complexity?

- **Make over-simplifications –> the ‘black-box approach’**
  - Based on standardized toxicity tests (e.g., OECD);
  - Consists of reducing information on toxicity with summarizing statistics (namely LCx/ECx) estimated at a target exposure duration;
  - Critical effect concentrations are specific to the environmental conditions.

❑ Nothing mechanistic;
❑ Nothing predictive;
❑ Current approach in ERA.

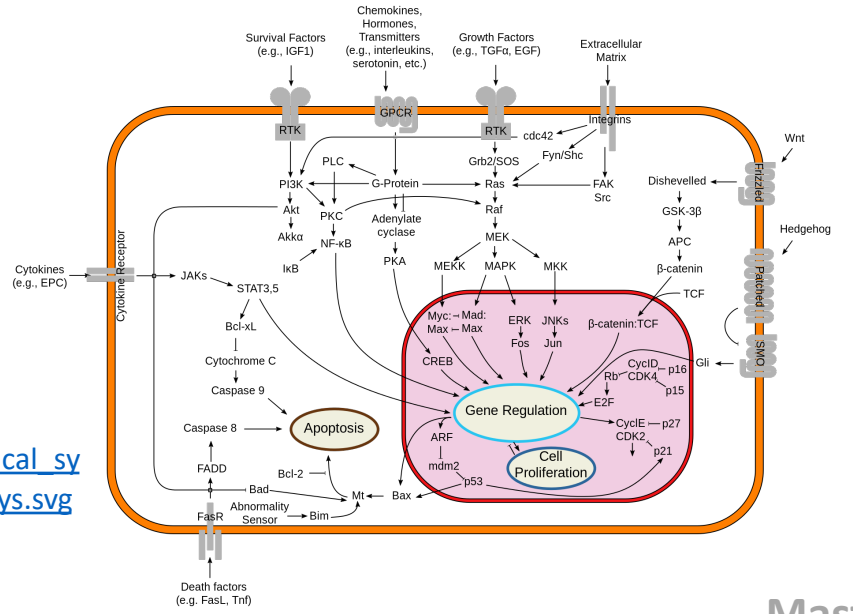


Fit of classical dose-response models

How to face with this complexity?

- Make over-simplifications –> the 'black-box approach'
- **Use a fully detailed model → the 'white-box approach'**
  - Deciphers the finest mechanistic aspects of chemical effects on organisms;
  - Consists of linking responses at the molecular, cellular and organ levels to the life-history traits.

❑ species- and compound-dependent;
❑ often over-parametrized models;
❑ highly data consuming.



https://en.wikipedia.org/wiki/Modelling_biological_systems#/media/File:Signal_transduction_pathways.svg

How to face with this complexity?

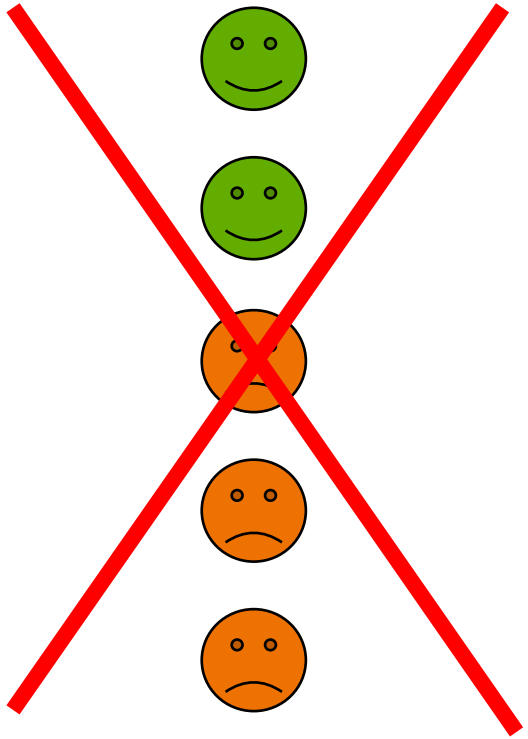
- Make over-simplifications –> the ‘black-box approach’
- Use a fully detailed model → the ‘white-box approach’
- **Make an idealization of the system → the ‘simple-box approach’**
  - **Understands** why toxic effects change over time, vary between species and toxicants, and depend on environmental conditions;
  - **Explains** links between life-history traits, as well as effects of chemicals over the entire life cycle (from egg to death);
  - **Predicts** effects under untested conditions;
  - Has parameters with a **physical/biological meaning;**
  - Remains as species- and compound-specific as possible.

  → Favours a ‘process-based’ also said ‘mechanistic’ approach

TKTD: the good compromise

- TKTD models allow describing and understanding **effects of chemical stressors** on organisms;
- They provide a general framework to quantitatively understand, and ultimately predict, the biological effects of chemicals **over time**;
- Chemicals are understood to be the chemical substances that are **not part** of the organism's "normal" functioning;
- Current TKTD models mainly focus on multicellular ectotherms or plants.

TKTD in comparison with other approaches

Features	NOEC/LOEC	DR models	TKTD models
Simplicity			
Model-independency			
Statistical correctness			
Tested concentration independency			
Target time independency			

- Chemicals need to be taken up into the body and transported to a target site before they will exert an effect;
- They may undergo biotransformation into other compounds (→ metabolites), which may be more or less toxic.
- They may be eliminated from the body (*e.g.*, dilution by growth).

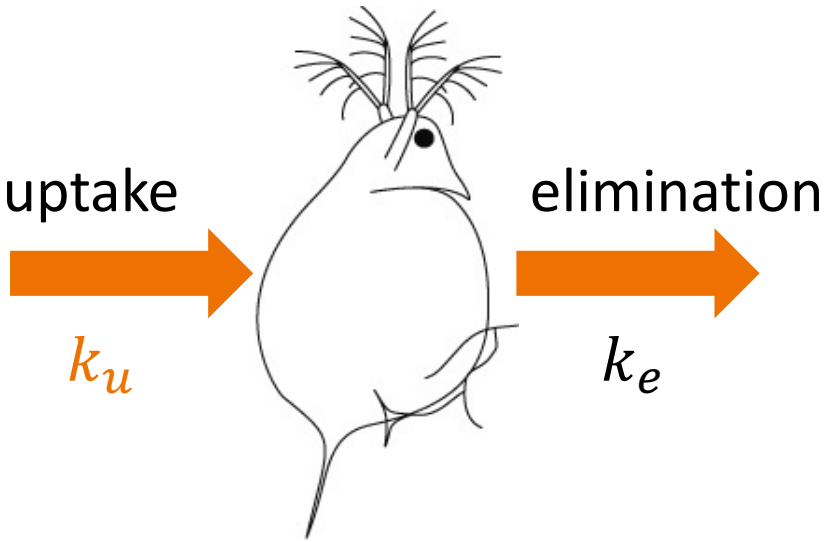
→TK models are ‘compartment’ models;

→One or more compartments;

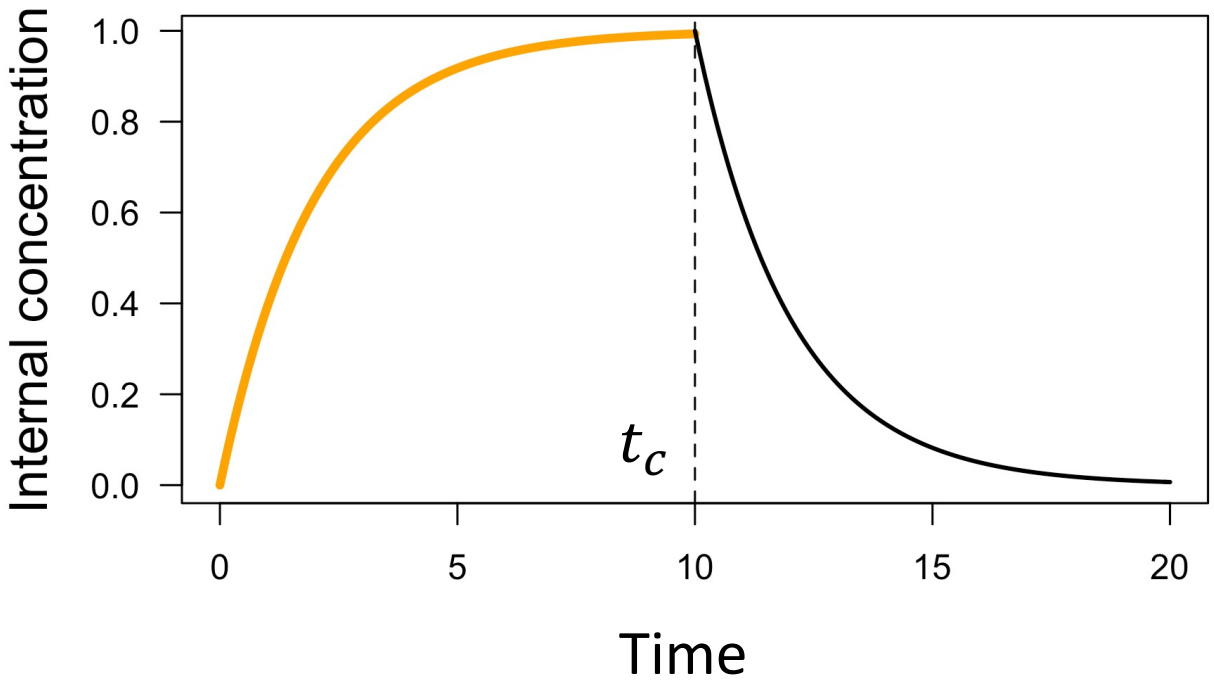
→The chemical is assumed to be evenly distributed within the compartment(s).

- **From very simplistic and general one-compartment model…**
- **… to models of intermediate complexity…**
- **… to very complex physiologically-based toxicokinetic models**

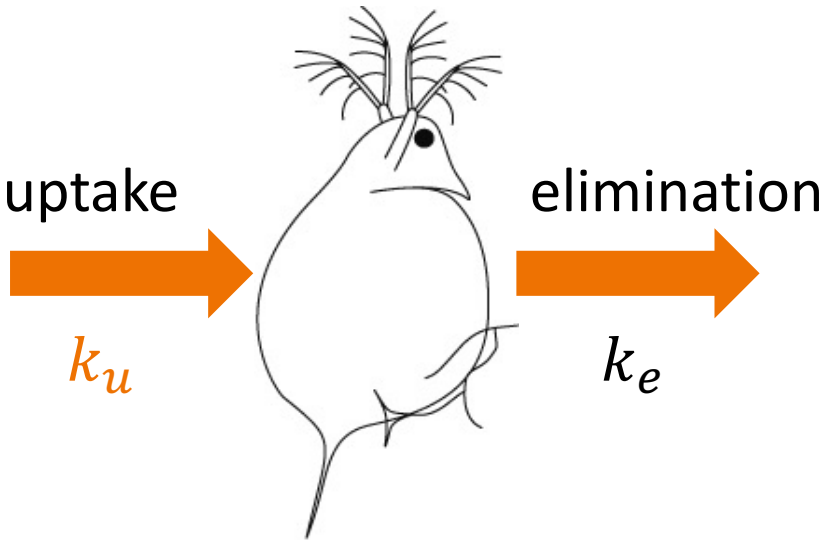
- **From very simplistic and general one-compartment model…**



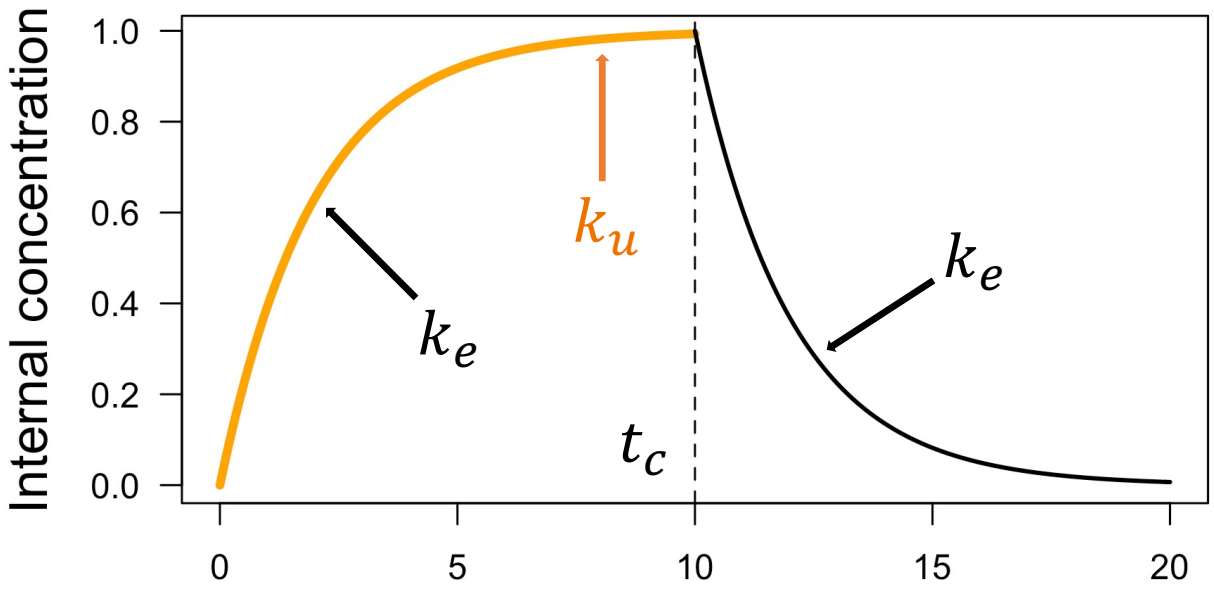
Bioaccumulation – Depuration curve



- **From very simplistic and general one-compartment model…**



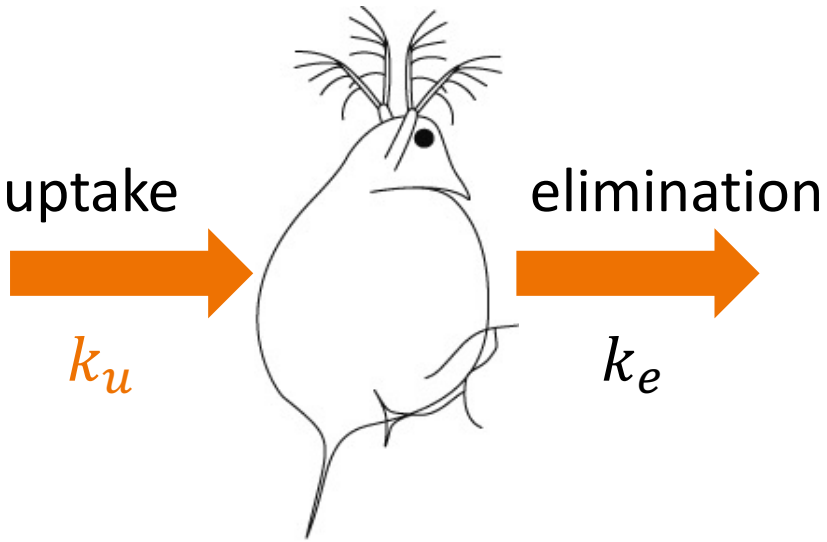
Bioaccumulation – Depuration curve



$$\begin{cases} \frac{dC_p(t)}{dt} = k_{uw} \times c_w - k_{ee} \times C_p(t) & \text{for } 0 \leq t \leq t_c \\ \frac{dC_p(t)}{dt} = -k_{ee} \times C_p(t) & \text{for } t > t_c \end{cases}$$

$$C_{obs,p}(t) \sim \mathcal{N}(C_p(t), \sigma^2_{C_p})$$

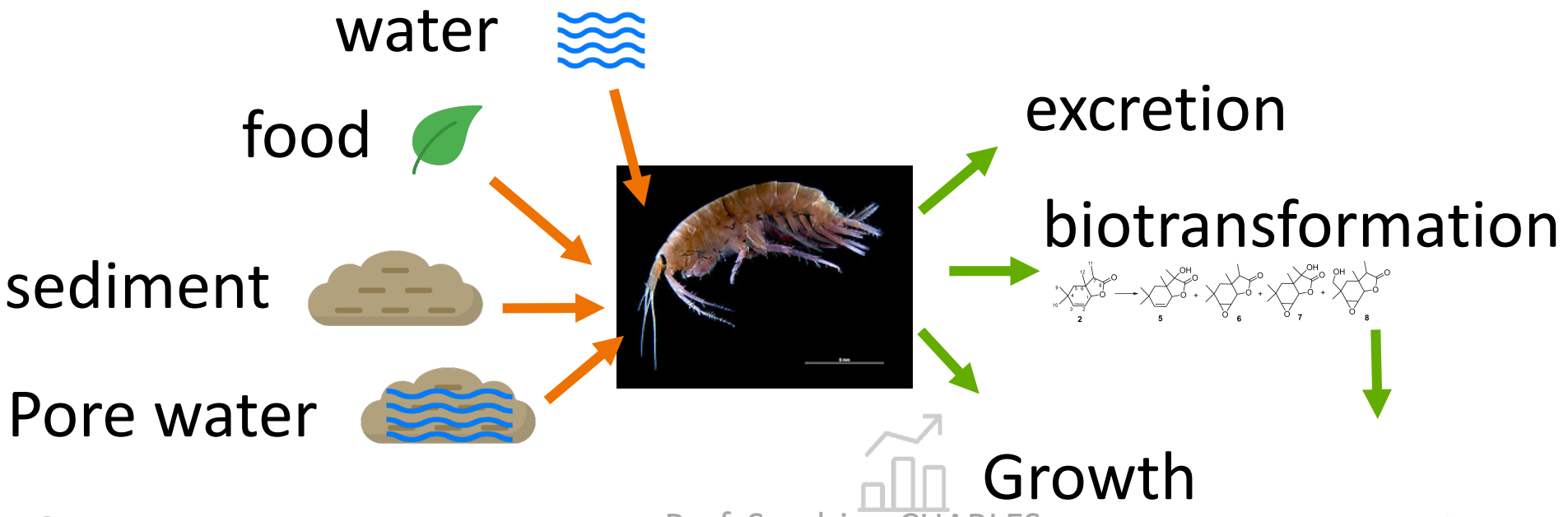
- **From very simplistic and general one-compartment model…**



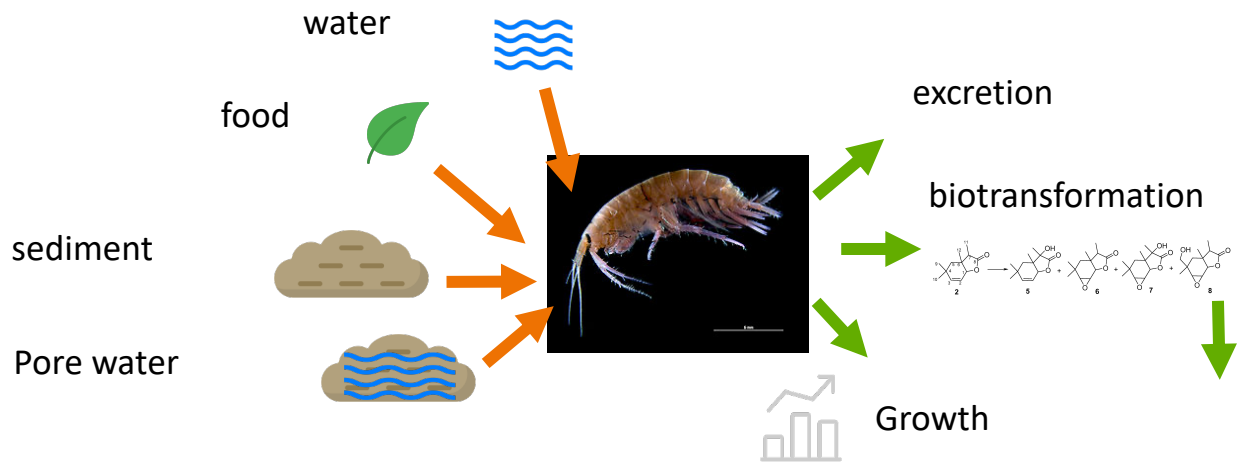
Bioaccumulation factor:

$$BCF_k = \frac{k_u}{k_e}$$

- From very simplistic and general one-compartment model…
- **… to models of intermediate complexity…**
  - TK models accounting for different sources and processes of accumulation and depuration on whole organism



Ratier et al. 2019. *Ecotoxicol. Environ. Saf.* 180: 33–42.



$$\begin{cases} \frac{dC_p(t)}{dt} = U - (E+M)C_p(t) \quad (1) \\ \frac{dC_{m_\ell}(t)}{dt} = k_{m_\ell} C_p(t) - k_{e_{m\ell}} C_{m_\ell}(t) \quad (2) \end{cases} \quad for \quad 0 \le t \le t_c$$

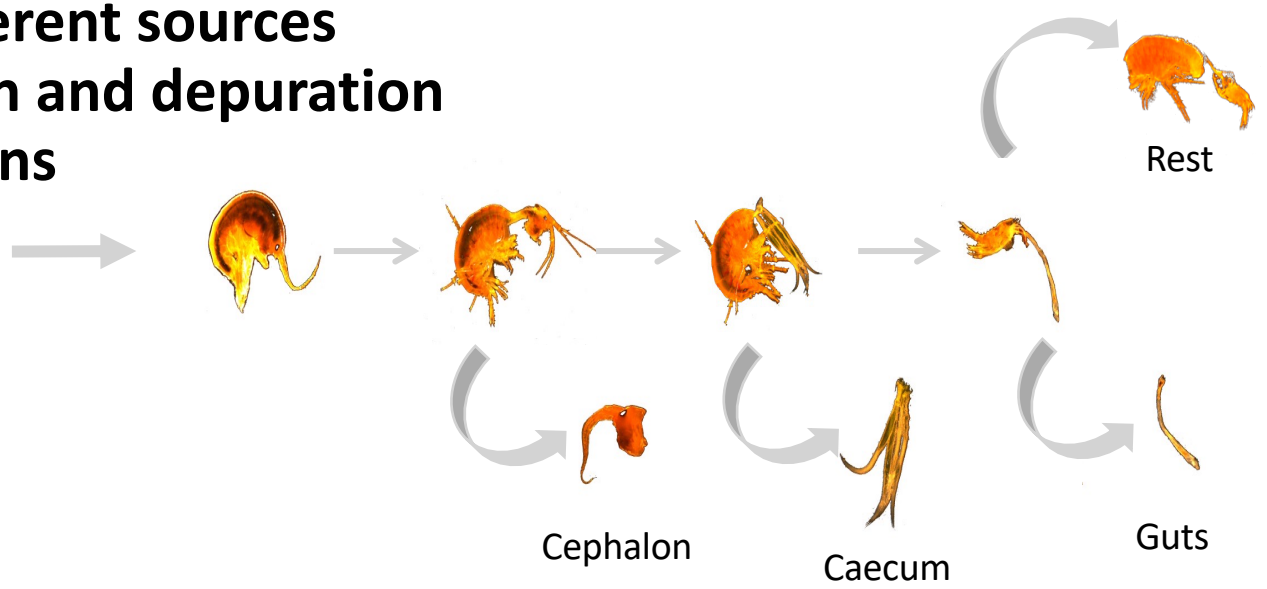
$$\begin{cases} \frac{dC_p(t)}{dt} = -(E+M)C_p(t) \quad (3) \\ \frac{dC_{m_\ell}(t)}{dt} = k_{m_\ell} C_p(t) - k_{e_{m\ell}} C_{m_\ell}(t) \quad (4) \end{cases} \quad for \quad t > t_c$$

$$C_{obs,p}(t) \sim \mathcal{N}(C_p(t), \sigma^2_{C_p})$$

$$C_{obs,m_\ell}(t) \sim \mathcal{N}(C_{m_\ell}(t), \sigma^2_{met_\ell})$$

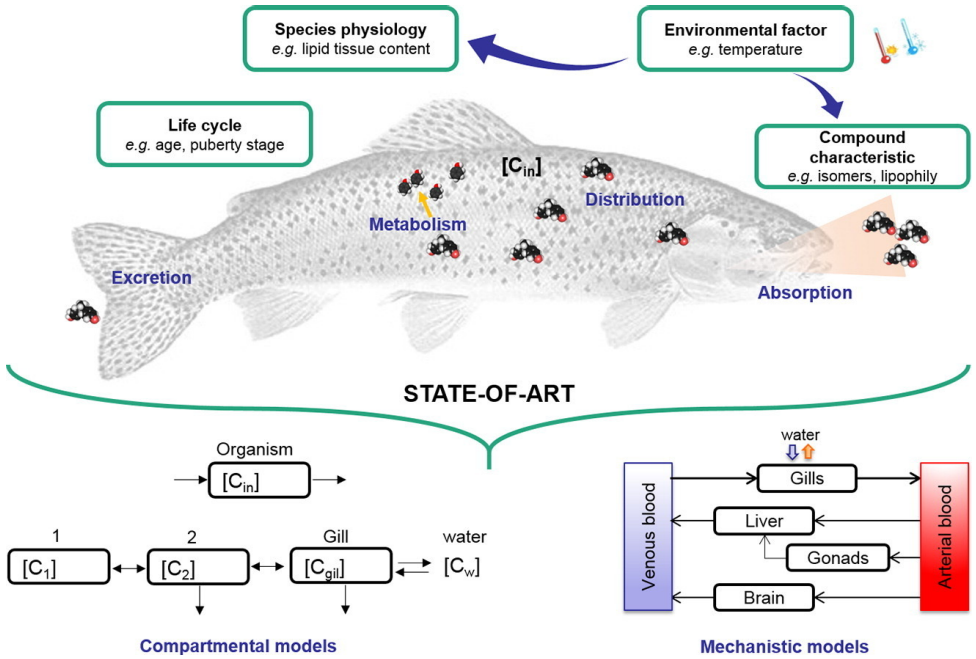
Ratier et al. 2019. *Ecotoxicol. Environ. Saf.* 180: 33–42.

- From very simplistic and general one-compartment model…
- … to models of intermediate complexity…
  - TK models accounting different sources and processes of accumulation and depuration on whole organism
  - **TK models accounting for different sources and processes of accumulation and depuration but differentiating target organs**



Gestin *et al*. 2020. *In preparation*.

- From very simplistic and general one-compartment model…
- … to models of intermediate complexity…
- **… to very complex physiologically-based toxicokinetic (PBTK) models**



Generally restricted to large-bodied organisms (*e.g.*, fish or mammals)

Grech *et al*. 2016. *Sci. Total Environ*. 578: 1–15.

MOSAIC Bioacc

127.0.0.1:4970

MOSAIC$_{bioacc}$

The MOSAIC$_{bioacc}$ application is a turn-key web tool providing bioaccumulation factors (BCF/BMF/BAF) from a toxicokinetic (TK) model fitted to accumulation-depuration data. It is designed to fulfil the requirements of regulators when examining applications for market authorization of active substances. Learn more

User Guide | Article | Video

Contact: sandrine.charles@univ-lyon1.fr

Beta version (updated on 29/09/2020)

This work is supported by the EUR H2O'Lyon (ANR-17-EURE-0018) of Université de Lyon (UdL), within the program "Investissements d'Avenir" operated by the French National Research Agency (ANR).

Measured concentrations

Please upload at least a four-column file in .txt or .csv format. The correct separator has to be chosen. Columns with headers, in exact order, must be the followings:

- The time points at which you measured concentrations (header = **'time'**)
- The measured concentrations within the organisms, must be in $\mu g.g^{-1}$ (header = **'conc'**)
- The exposure concentration in water, sediment, food and/or pore water, nominal or measured (constant over time), must be in $\mu g.mL^{-1}$ or $\mu g.g^{-1}$ (header = **'expw'**, **'exps'**, **'expf'** or **'exppw'**)
- The IDs of the replicates (header = **'replicate'**)

Other columns can be added in the file:

https://mosaic.univ-lyon1.fr/bioacc

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- The IDs of the replicates (header = **'replicate'**)

Other columns can be added in the file:

Sub-lethal endpoints

- **The Dynamic Energy Budget (DEB) theory**
  = a unified approach to deal with energy allocation by organisms, an integrated view of an organism as a dynamical system
  → The standard DEB model for animals

- **The DEBtox equations**
  = a way to include the energy-budget approach in the TD module
  → Effects on growth and reproduction involve a change in DEB parameters

http://www.debtox.info/

Lethal endpoints

The **G**eneral **U**nified **T**hreshold model of **S**urvival

GUTS

What is GUTS?

ENVIRONMENTAL Science & Technology

CRITICAL REVIEW

2011, 45, 2529–2540

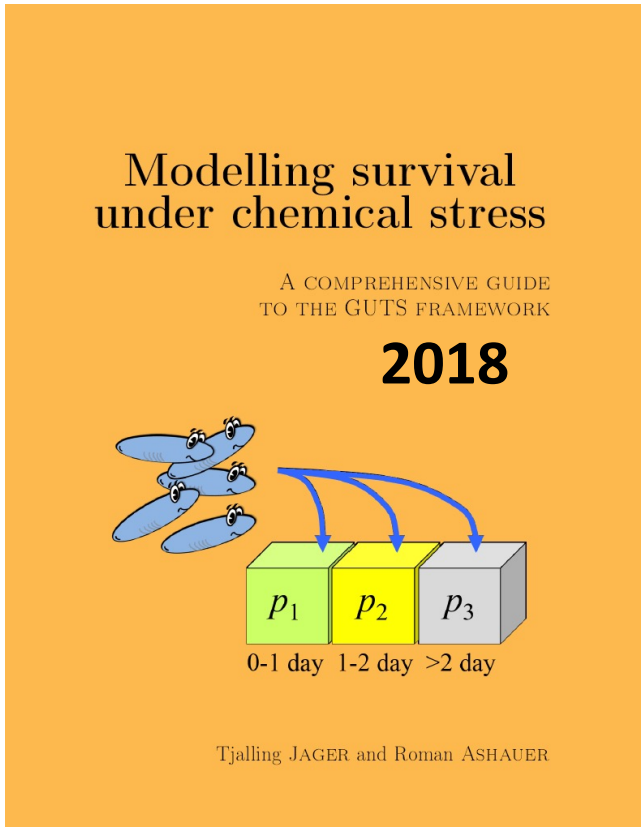
pubs.acs.org/est

General Unified Threshold Model of Survival - a Toxicokinetic-Toxicodynamic Framework for Ecotoxicology

Tjalling Jager,[†] Carlo Albert,[‡] Thomas G. Preuss,[§] and Roman Ashauer[‡,*]

→ A theoretical framework about stressors effects on survival over time, based on hypotheses related to:

- Stressor quantification (choice of a dose metric);
- Compensatory processes;
- Nature of the death process.



TKTD models for Environmental Risk Assessment

Scientific Opinion on the state of the art of Toxicokinetic/Toxicodynamic (TKTD) effect models for regulatory risk assessment of pesticides for aquatic organisms

efsa
European Food Safety Authority

ej EFSA Journal

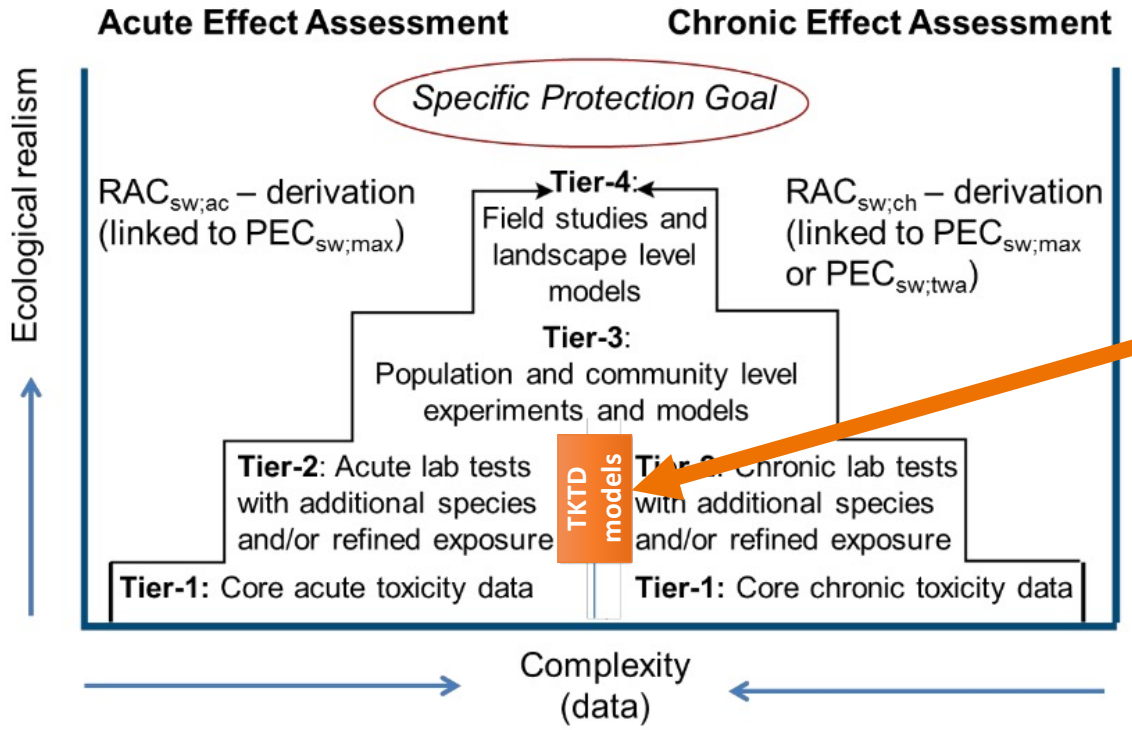
https://efsa.onlinelibrary.wiley.com/doi/full/10.2903/j.efsa.2018.5125#

TKTD models for Environmental Risk Assessment

Scientific Opinion on the st… t of Toxicokinetic/Tox… D) effect model… isk assessment of … for aquatic organisms

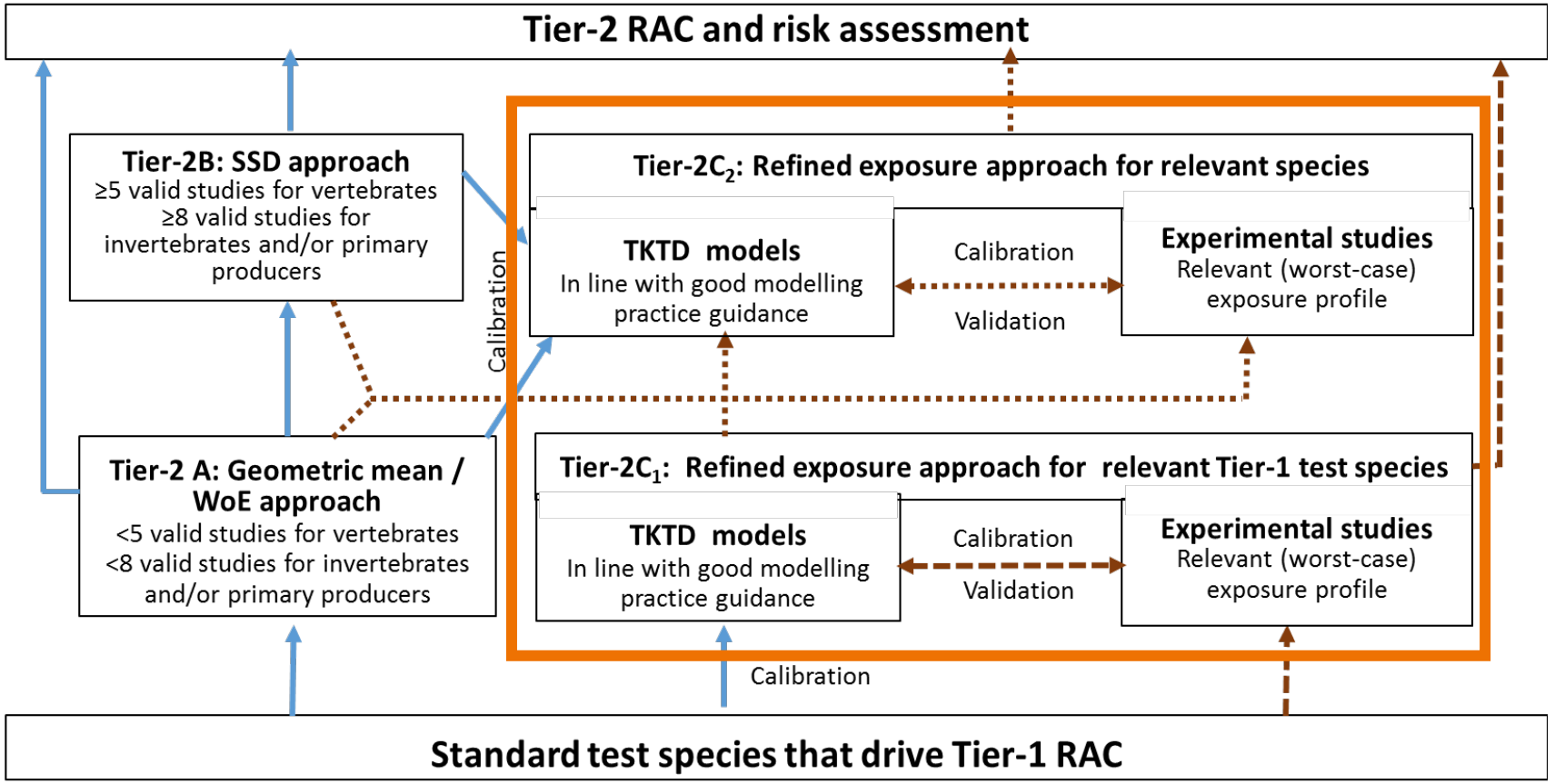
APPROVED 27-06-2018

https://efsa.onlinelibrary.wiley.com/doi/full/10.2903/j.efsa.2018.5125#



European Food Safety Authority

TKTD models in Tier-2C



Solid blue lines → **Standardised exposure**:
Experimental studies with standard and/or additional test species and exposure conditions in line with Tier-1 tests (worst-case approach)

Broken lines ⇢ **Refined exposure (Tier-2C$_1$):**
⋯▸ **Refined exposure (Tier-2C$_2$):**
Tests with standard and/or additional species and refined exposure conditions informed by predicted field exposure profiles

TKTD models in Tier-2C

- TKTD modelling may be used to address (the threshold for) individual-level effects occurring from **time-variable exposure regimes** on aquatic vertebrates and invertebrates (Tier-2C), **even if TKTD models could also be used from data collected at Tier-1**;
- The **GUTS** framework is appropriate as it is for lethal effects in the acute and the chronic risk assessment scheme;
- The **DEBtox** framework is promising and seen as very relevant for sub-lethal effects in the chronic risk assessment scheme;
- **Plant models** need further standardisation, documentation, calibration and validation, except for *Lemna minor* which is ready-to-use.

1 **Calibration**

Fit a TKTD model on toxicity test data and get parameter estimates associated with their uncertainty

2 **Validation**

Simulate an effect over time under a time-variable exposure profile and compare with observed data from a refined toxicity test

→ Three validation criteria are recommended by EFSA

3 **Prediction**

Make simulations under realistic scenarios to assess risk on how far is the exposure profile from causing a pre-defined effect.

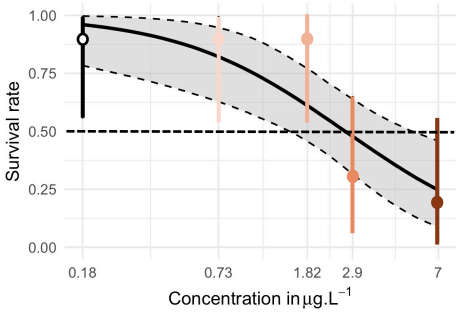
Using GUTS models → Concept of the x% Lethal Profile (LPx) = Multiplication Factor leading to an additional x% reduction in the final survival rate.

efsa workflow for ERA

European Food Safety Authority

TKTD models as new tools for Environmental Risk Assessment

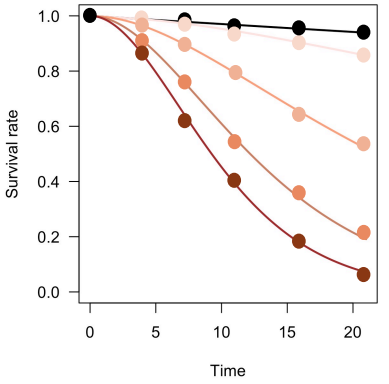
Compare the classical LC_{50} value at final time with the corresponding calculations from GUTS-IT and GUTS-SD models



- **What do we expect**?

  Because TKTD models use all data over time at each tested concentration, we expect a better precision (i.e., a reduced uncertainty) of the LC_{50} calculated with GUTS models

- **20 standard survival datasets**:
  - 10 species / One toxicant (**Chlorpyriphos**)
  - One species (**D. magna**) / 7 toxicants
  - Three other datasets



Bayesian implementation workflow

- Fit to each dataset
  - IT • **Model GUTS-IT**
  - SD • **Model GUTS-SD**
  - C • **Classical dose-response model**

Package 'morse' 3.1.0
[Baudrot et al., 2018]

https://CRAN.R-project.org/package=morse

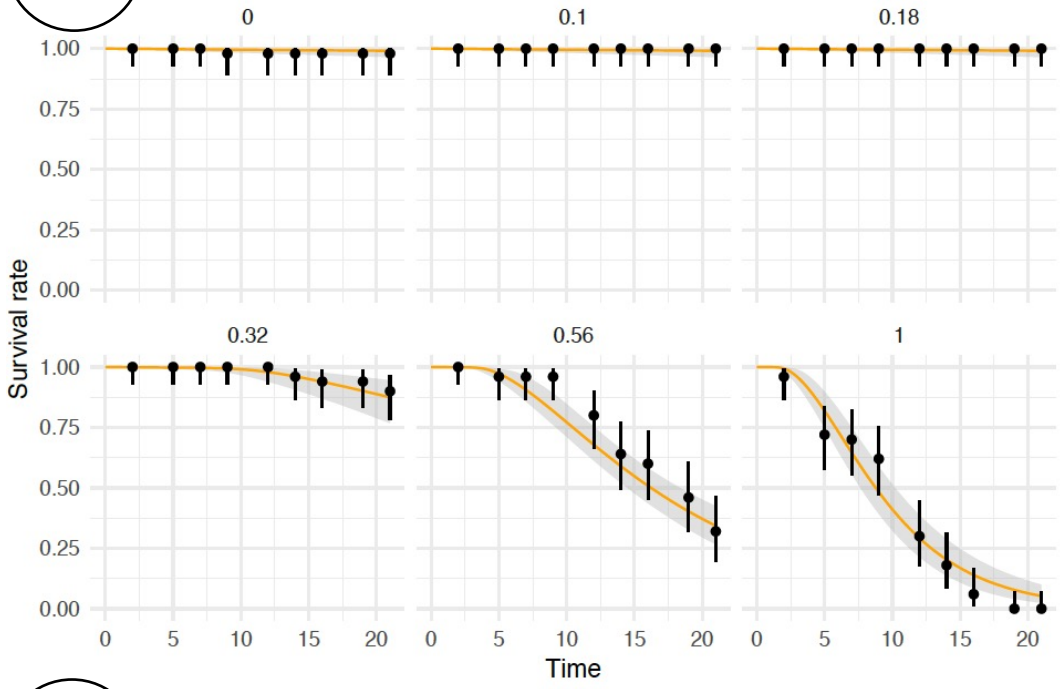
- Get parameter estimates
  - Visual fit quality
  - Goodness-of-fit criteria
- LCx,t calculations
  - x = 50%
  - t = final time

http://pbil.univ-lyon1.fr/software/mosaic/guts
[Charles et al., 2017]
[Baudrot et al., 2018]

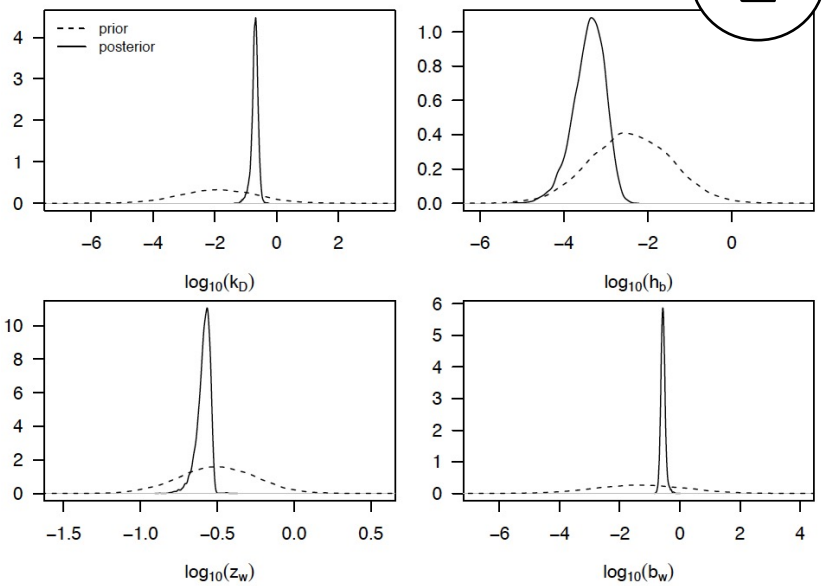
D. magna / Potassium dichromate

GUTS-SD model

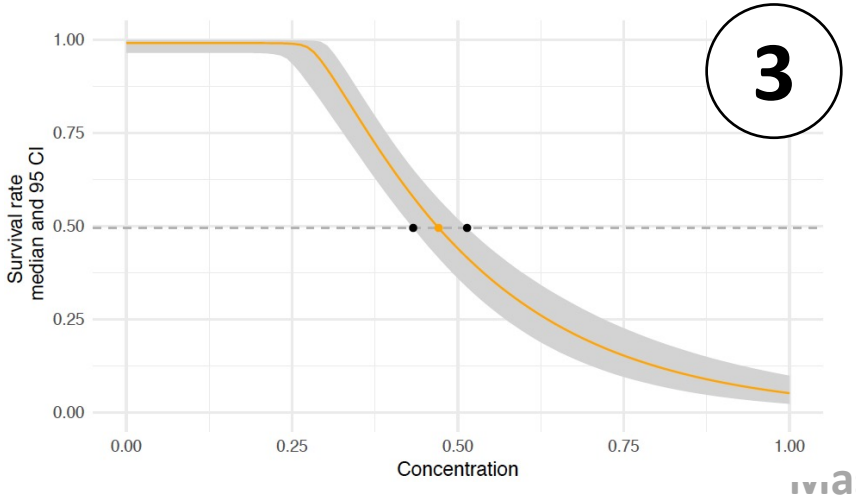
1 Visual check of fit



2 Parameter estimates



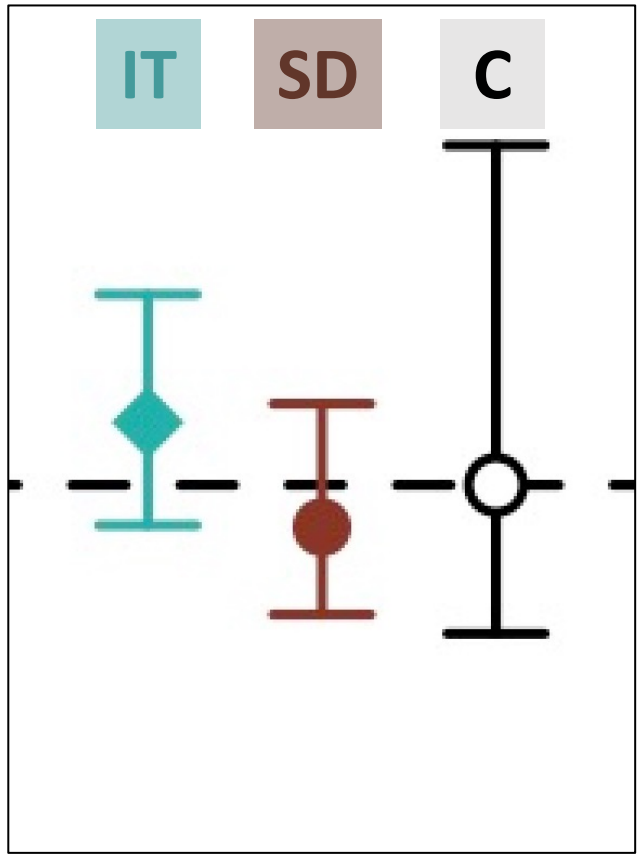
3 Predicted dose-response curve at day 21



4 LC_{50} calculation

$$LC_{50} = 0.47 \quad [0.43 ; 0.51]$$

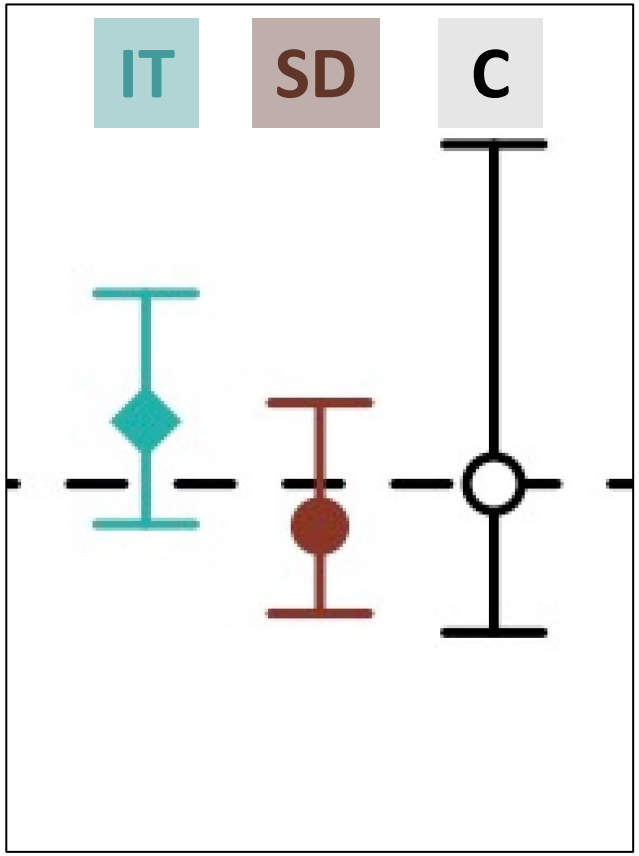
Comparison of LC_{50} calculations



***D. magna* / Potassium dichromate**

Reference value = LC_{50} median value obtained with the classical dose-response model (**C**)

Comparison of LC_{50} calculations

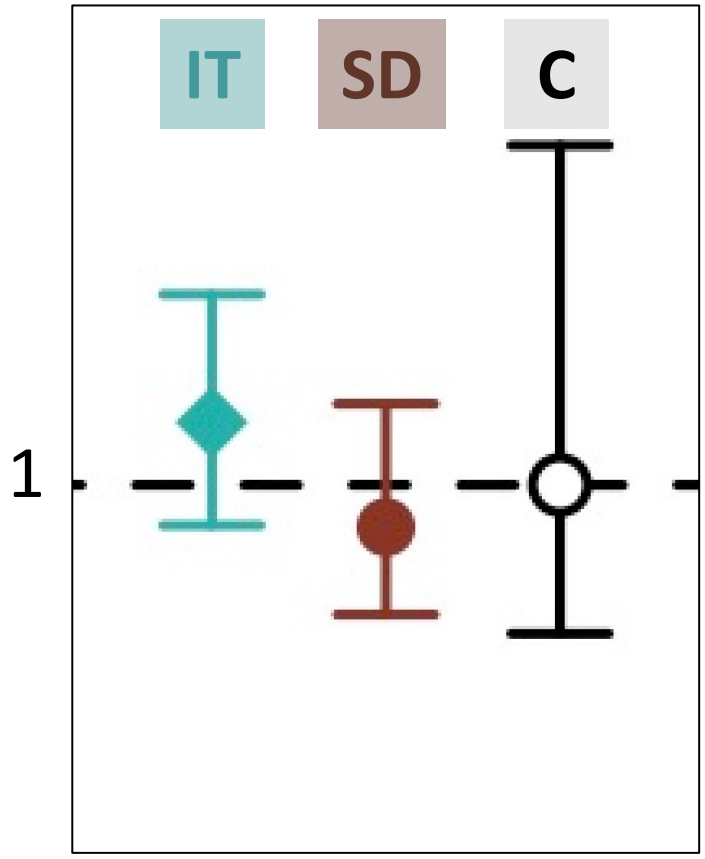


***D. magna* / Potassium dichromate**

As expected, we get a better precision with GUTS models

Reference value = LC_{50} median value obtained with the classical dose-response model (**C**)

Comparison of LC_{50} calculations



***D. magna* / Potassium dichromate**

- **Three questions arise:**

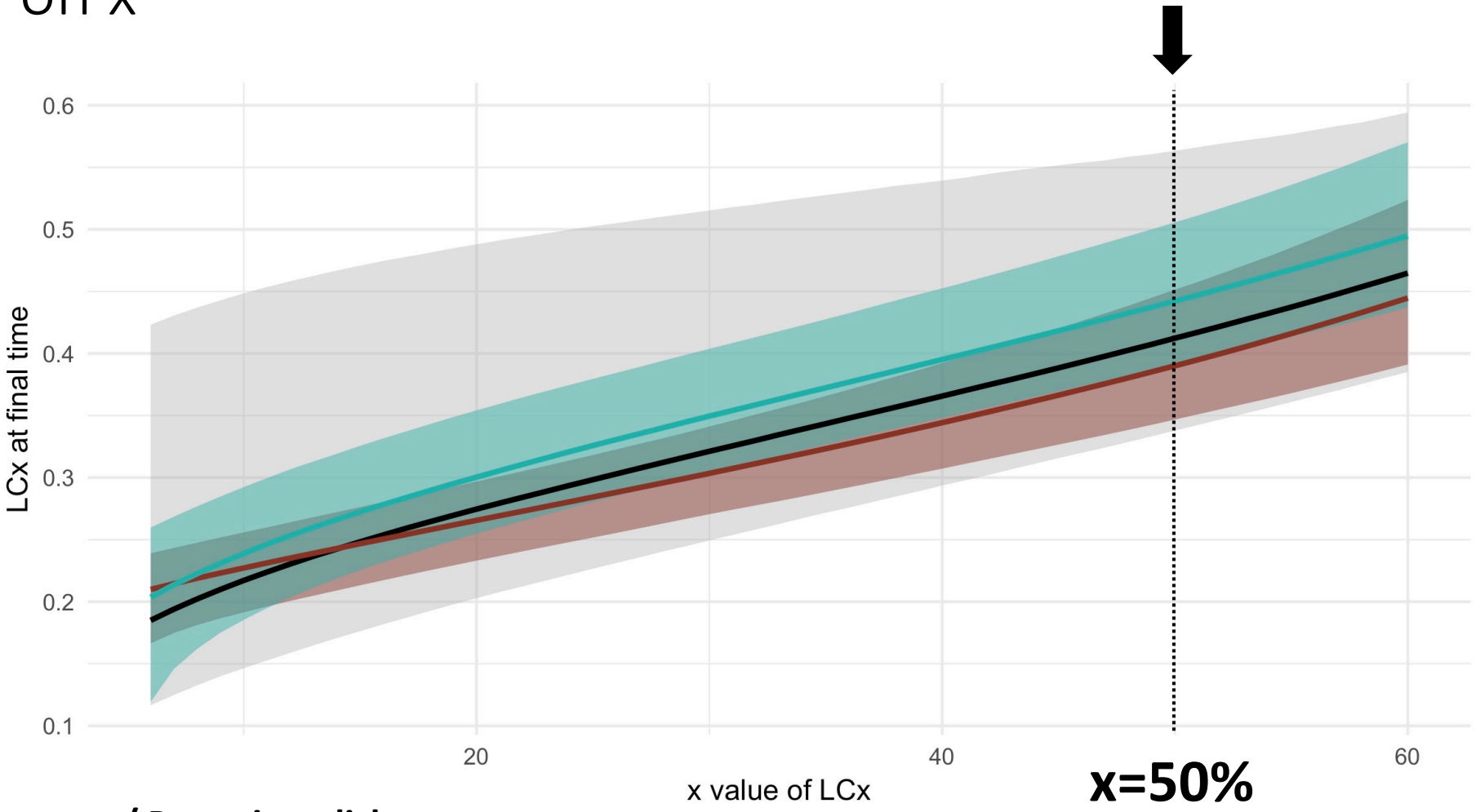
1. Does the better precision depend on x?
2. Does the better precision depend on the exposure duration?
3. Does the better precision depend on the dataset?
   - On the species?
   - On the toxicant?

One dataset: LCx at final time versus x
→ The better precision does not depend on x

IT

SD

C



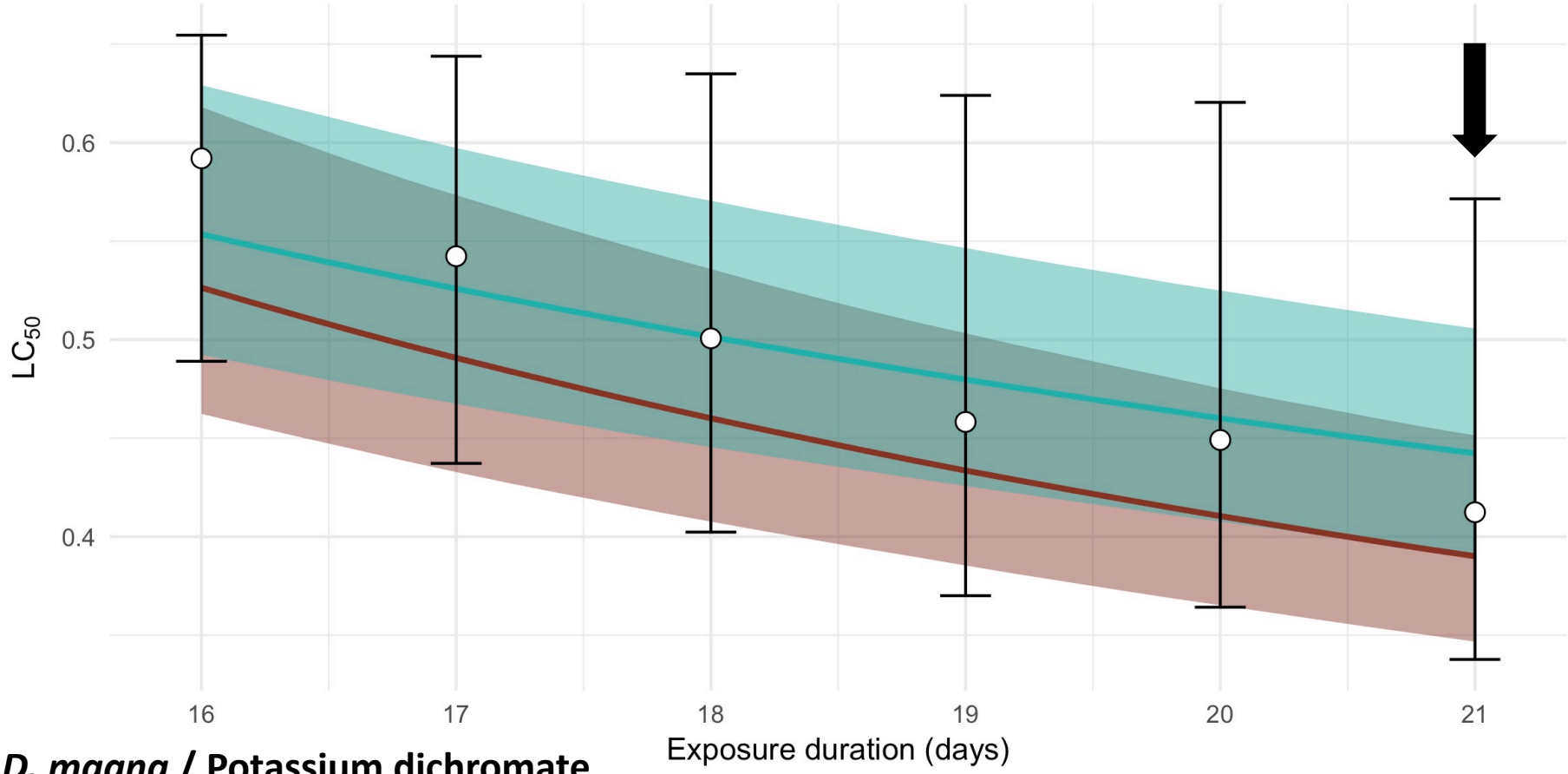
***D. magna* / Potassium dichromate**

IT

SD

C

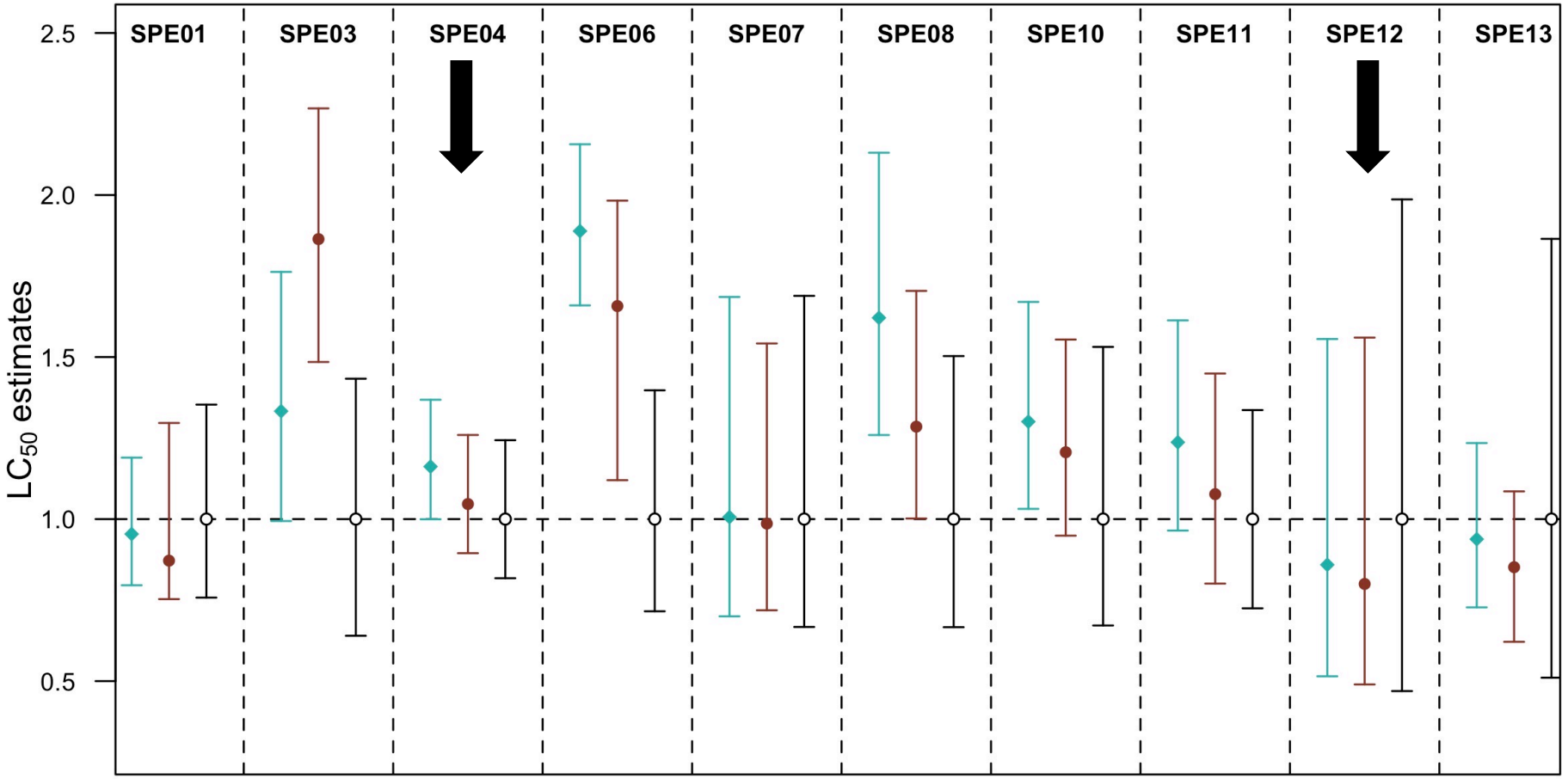
One dataset: LC_{50} over time
→ The better precision does not depend on the exposure duration



***D. magna* / Potassium dichromate**

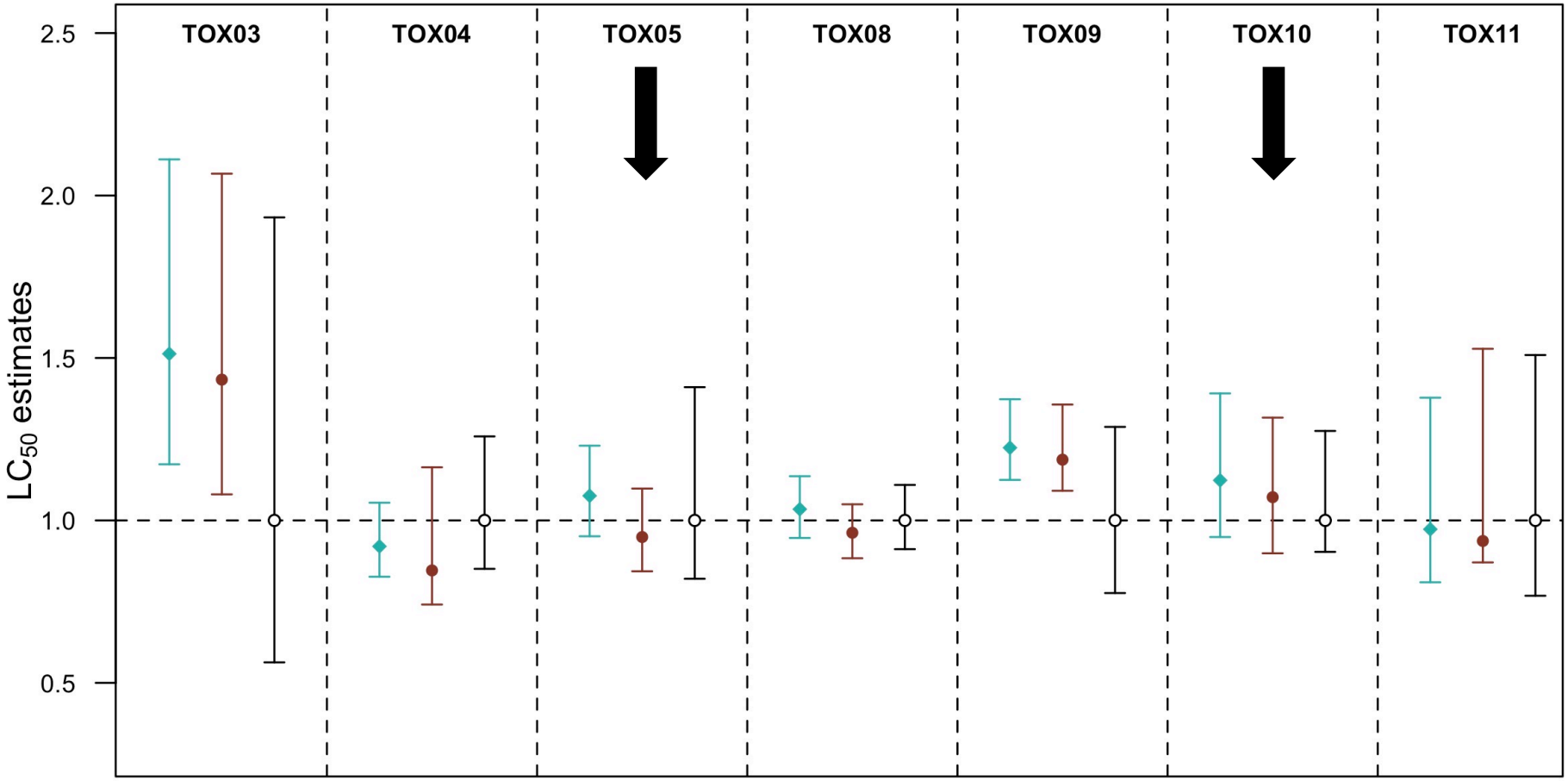
10 species / One toxicant: LC_{50} at final time
→ The precision depends on the species

IT
SD
C



One species / 7 toxicants: LC_{50} at final time
→ The precision depends on the toxicant

IT
SD
C



Conclusion

- TKTD models account for all the data from the beginning to the end of the experiment: **no data are lost**;
- GUTS models allow the estimation of the **LCx,t whatever x and whatever t**, even at time points not in the experimental design;
- Even if dependent on the dataset, for most of them, GUTS models provide LCx,t estimates with a **better precision** compared to the classical dose-response one;
- GUTS models can **easily** be fitted on any dataset either on-line (MOSAIC platform) or with the R software (package 'morse').

http://pbil.univ-lyon1.fr/software/mosaic/guts

https://CRAN.R-project.org/package=morse