Evaluating Toxicokinetic Models as Predictors of Internal Concentration in Environmentally Relevant Species

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1. Background

1. The use of ***in silico*** and ***in vitro*** methods, commonly referred to as **New Approach Methodologies (NAMs)**[1], have been increasingly proposed as a mean to support environmental safety decisions for chemicals and **ensure environmental protection**.
2. Toxicokinetic (TK) modelling can provide reliable predictive capabilities offering opportunities to **replace animal use** and **increase efficiency** in terms of **time / cost**[2] and **mechanistic understanding** of effects.
3. *Daphnia magna* is an aquatic invertebrate species, used extensively as a model organism in ecotoxicology and safety assessments. However, **quantitative TK data** for invertebrates including *Daphnia magna* are limited, resulting in a lack of robust TK models. Current *D. magna* TK studies are **chemical specific**, with restricted application to other chemicals.
4. Here we apply the Arnot & Gobas[3] proposed two-compartment model describing the exchange of a chemical between the external water and the organism inhabiting it over a given exposure time and evaluated its performance as a general predictor.

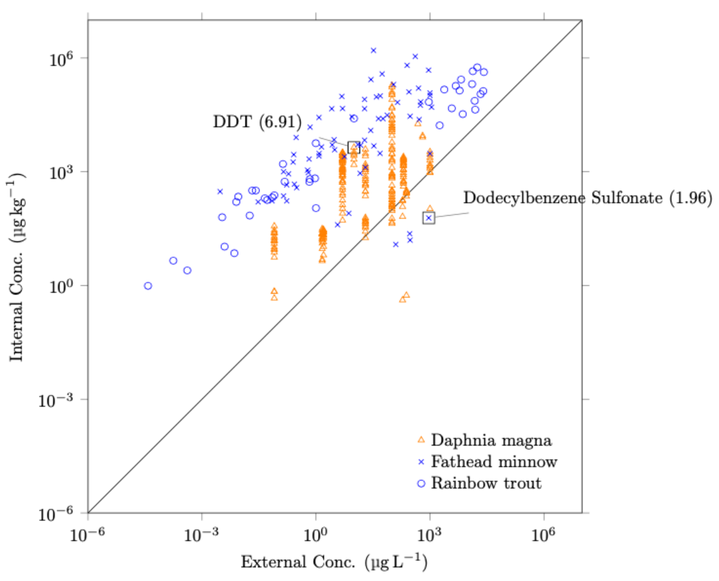
2. Aim

The aim of this study was to **develop a general cross-species two-compartment model** to predict the internal concentration of species based on existing TK modelling concepts and to evaluate the model using existing data from the literature. The predictive performance in daphnia is benchmarked against a relevant fish species dataset (fathead minnow and rainbow trout).

4. Results – Literature data review

- Figure 1 shows that most chemicals in the dataset have internal concentrations that exceed the studies external concentration (points on the vertical axis above the bisection).
- Two long exposure datapoints highlight the effectiveness of the Octanol-water partition coefficient on the **internal concentration** with a two-day DDT assay on daphnia with a high log-Octanol Water Partition coefficient of **6.91** having a higher internal concentration compared to a 32-day fathead minnow dodecylbenzene sulfonate assay, with a low log-Octanol water partition coefficient of **1.96**.

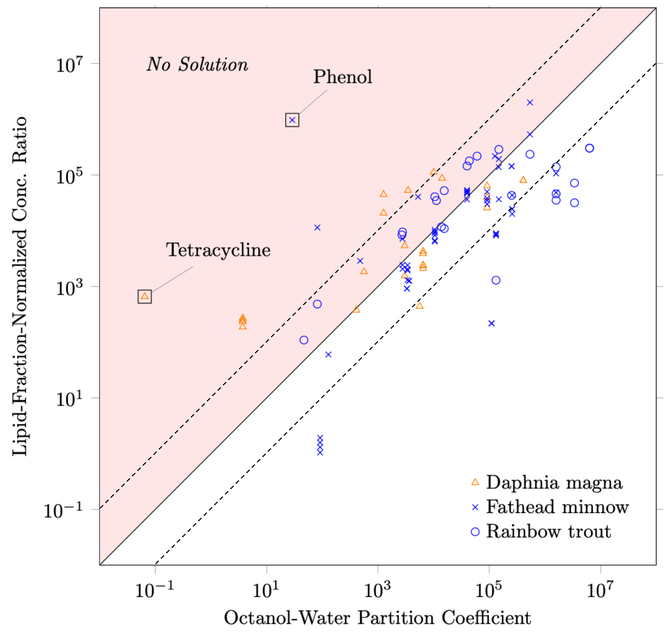
Figure 1



5. Results – Model predictions

- The model prediction performance is independent of species (Figure 2).
- ~70% Predictions are within 1 order of magnitude.
- Phenol & tetracycline are **outliers** potentially caused by ionisation of the chemicals.

Figure 2



3. Methodology

D. magna TK data extraction from the literature

1. Studies that measured the external and internal concentration of at least one chemical over a range of exposure times were considered. Due to lack of readily available data a digitizer was used to extract TK data from graphical plots.

Data standardisation and combination

2. The external and internal concentration, wet weight and exposure time were all standardized and were then combined with the Stadnicka[4] fish dataset containing similar data for the fathead minnow and rainbow trout.

Performance evaluation framework for cross-species comparison

3. A key modelling insight from Arnot & Gobas[3] assumes that for negligible metabolic elimination, and for long exposures, the internal / external concentration ratio (k_r) is predicted by the lipid fraction (v) and the octanol-water partition coefficient ($K_{o/w}$). The novelty of this work is using this insight to predict the internal concentration across multiple species with minimum data requirements.

$$k_r = v \cdot K_{o/w}$$

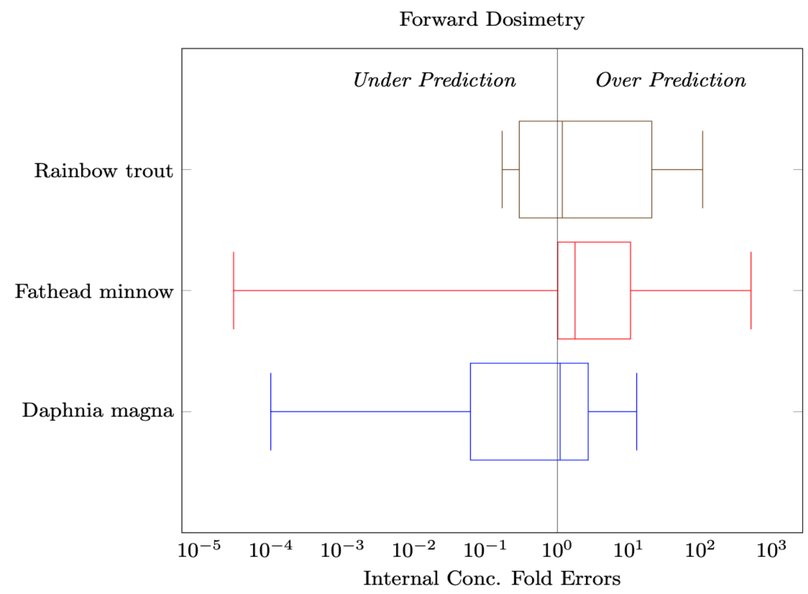
Steady state output data

4. The state of the system when the internal concentration measurement was taken is required. In practice, the system reaches an effective steady- state when the rate of chemical exchange is sufficiently small, which was shown to be 100 µg kg^{-1} d^{-1}. 80% of datapoints for daphnia internal concentration measurements did not reach steady state, due to the time course nature of the measurements.

6. Results – Model performance

- Predicted versus measured **internal concentrations** for daphnia have a **median fold-error of 1**, while fathead minnow and rainbow trout median fold-errors over-predict (Figure 3).
- Error dispersion measured by inter-quartile range is **smaller** in daphnia than in rainbow trout, but **larger** than fathead minnow.
- Rainbow trout has the **smallest prediction range** while fathead minnow has the **greatest.** Both daphnia and fathead minnows prediction span is biased towards **under-prediction**.

Figure 3



7. Take home messages

- Successfully **filled quantitative TK data gaps** for daphnia and provided this in an **available R package** (*TKAquatic*) for use by the research community, which is essential for *in silico* modelling and the implementation of high-throughout NAMs for environmental risk assessment.
- Developed a **novel proof of concept** performance evaluation framework for cross-species comparison highlighting the ability to generate **robust predictions** in aquatic organisms with **minimum data requirements**.
- On average predictions in **daphnia** have been shown to be **most reliable**.

[1]Kavlock, R. et al. (2018). Accelerating the pace of chemical risk assessment. Chemical research in toxicology, 31(5), 287-290.
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[3]Arnot, J. A., & Gobas, F. A. (2004). A food web bioaccumulation model for organic chemicals in aquatic ecosystems. Environmental Toxicology and Chemistry: An International Journal, 23(10), 2343-2355.
[4]Stadnicka, J., et al. (2012). Predicting concentrations of organic chemicals in fish by using toxicokinetic models. Environmental science & technology, 46(6), 3273-3280.

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