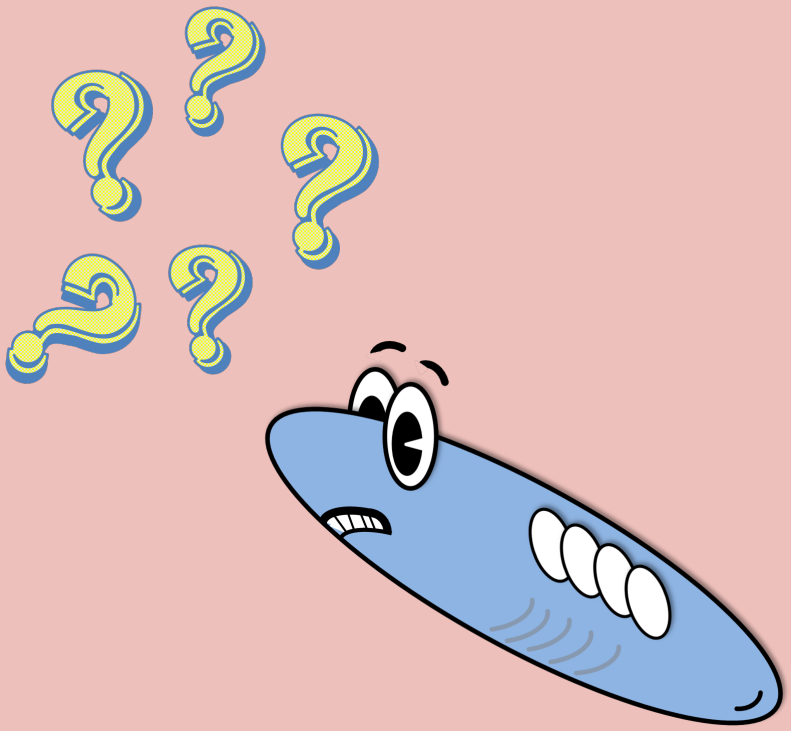
Big Open Questions in TKTD modelling

(and some not so big ones)



Tjalling Jager

Big open questions in TKTD modelling

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Contents

Preface

About this book

I have been working on TKTD models since 2002, first as a member of the department of Theoretical Biology of the VU university in Amsterdam, and since 2015 as a self-employed researcher at DEBtox Research. Over the years, I realised that there were many things that I wanted (or needed) to know that were still unknown. Furthermore, I realised that answering these questions was, in many cases, not easy at all (especially for a theoretician with no access to a lab). I have always happily shared these questions, and my thoughts on how to address them, with anyone wanting to listen, and these thoughts also made it into my e-books and publications. Here they are bundled together to, hopefully, inspire a new generation of ecotoxicologists to address these fundamental scientific questions about the mechanisms underlying the response to toxicants (and some practical issues as well).

This collection of questions is, at the moment, a very personal one, and certainly not exhaustive. These are the questions that I see as important and interesting, and many of the citations are to my own work.[1] However, TKTD modelling is not a finished subject, and neither is this book. I expect to add subjects (and hopefully citations to new and exciting research) for a while. I am open to contributions from the reader, both additional questions and additional references to potentially useful approaches. So, please email me if you want to see/make an addition, and I will consider it seriously.

It would be good if there was some kind of platform for discussing TKTD models and the needs for further research. In my experience, it's not so easy to get everyone on board and committed to discussion. Nevertheless, a closer collaboration (or at least more communication) between TKTD scientists could lead to new ideas and possibly avoid modelling failures and unnecessary overlap. Therefore, if you see an opportunity to facilitate discussion in the TKTD community, please go for it!

Who is this book for?

This book is intended for TKTD modellers that need some inspiration for developing new projects. It could also be interesting for potential research funders, if they are willing to focus on issues that are less 'trendy' but very much needed to progress the field. Some of the topics in this book require serious effort, serious funding, and serious expertise. These probably require a consortium to address. However, on other topics, a student's project could already make meaningful headway.

To fully appreciate the questions in this book, you should have a firm grip on the concepts of TKTD modelling in general, and the concepts underlying GUTS and DEBtox in par-

[1] For most of the paywalled papers where I am first author, an 'author version' is available for free download. See my publication list at https://www.debtox.nl/downloads/publication_list.pdf, which includes the DOIs and the links to the free versions. Or email me for an official PDF.

ticular. These topics are only very briefly introduced in this book; much more thorough introductions are provided in my other e-books (see my author page on Leanpub for a full list of books: https://leanpub.com/u/tjallingjager). This book is (for now) math-free; the references provided will have those details. I guess this is not a book that you would want to read from front to back; it is probably more for browsing and selecting particular sections to read.

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Chapter 1

Introduction

Toxicokinetic-toxicodynamic (TKTD) models provide a mechanistic link between the external concentration (as function of time) that an organism is exposed to, and the observed toxic effects, as they develop over time [1, 8]. As the name already implies: TKTD models are built from a toxicokinetic module, whose output forms the input for a toxicodynamic module (Figure 1.1).

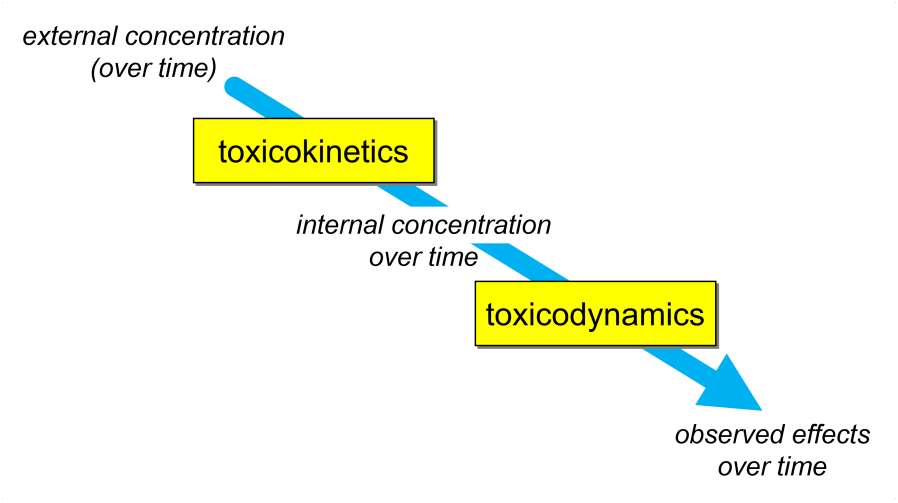

Figure 1.1: General scheme for TKTD models. This figure from [5].

The advantages of TKTD models are now widely acknowledged, and these models have undergone quite a surge of application and development since the early 2000's.[1] They have been included into guidance from OECD [11] and ISO [4], and more recently into EFSA opinions and guidance for risk assessment purposes [3, 2]. These developments might give the impression that TKTD modelling is a mature field where all models are 'finished works'. I would say this is incorrect; we do currently have some very useful practical models available, but many questions remain, and many of them are quite fundamental. In this book, I want to list, and briefly discuss, the topics that I see as important for in-depth research. I focus on GUTS and DEB-based TKTD models (or DEBtox, in the broad sense). These are the only TKTD approaches for animals with a substantial track record, and the only ones that were so far considered for risk assessment (for animals), as far as I know. TKTD models have

[1] See lists of publications at https://www.debtox.info/papers_survival.html and https://www.debtox.info/papers_debtox.html.

also been developed for plants (e.g., [10, 12]), however, I will not deal with those (for the moment) as I don't have a lot of experience with them.

As you can see from the GUTS and DEBtox studies that have been published, the focus has moved towards model application, rather than model evaluation, improving scientific understanding, and further model development. Even when there is model development, it is usually aimed at risk assessment, either trying to improve the assessment or trying to avoid the costs for additional experimental testing. This reflects the funding of this research, which currently mainly comes from chemical industry and organisations like EFSA, with a strong focus on risk assessment. It probably also reflects a deeper truth about the field of ecotoxicology, which has traditionally grown in response to real-world pollution problems, rather than driven by the desire to develop (quantitative) theory. When there are new developments, it is most common that these are supported by existing data sets, produced for a very different purpose, which is obviously not optimal for addressing fundamental TKTD questions.

In general, I would say that, at this moment, we have sufficient theory, ideas and models, but we need targeted and structural experimental effort to test these ideas and to select the most relevant ones for future updates of GUTS and DEB-based TKTD modelling (or dream up entirely new frameworks). Or, in any case, to elucidate the strengths and limitations of the models. Standard toxicity tests, or existing data sets, are not going to be enough: experimental effort needs to be structured around the particular TKTD question. The topic of experimental testing is discussed further in Section 8.1.

A disturbing (for me) trend in ecotoxicology is, nowadays, the drive to use artificial intelligence or machine learning (or whatever automated large-data statistical approach), attempting to extract useful information from large amounts of (old) data. While this is potentially useful to identify general patterns, or have more efficient interpolation of the data, I don't see this as a fruitful pathway to improve our understanding of the world around us (which is what science is all about, after all). In my opinion, we don't need more AI, we need more HI (human intelligence). To be clear, I don't see TKTD models primarily as tools for risk assessment. The intention should be, in my opinion, to develop mechanistic theory on how chemicals affect organisms. This theory can then be translated into models, and those models tested on real-world observations. The resulting fit or misfit provides information on the usefulness of our theory. As a spin-off, models will appear that can be used for practical application, but I don't see that as the main driver. For more discussion on the role of modelling in environmental/biological science, I like to refer the reader elsewhere [7, 9]. However, I do want to stress here that the point of modelling is not to get a curve through your data (there's better ways to do that). A good fit of a model is meaningless when the model is not mechanistic [6].[2]

In the subsequent chapters, I will highlight a number of questions that require deep HI and (in most cases) dedicated and detailed experimental work. Many of these questions are related to each other; it may be hard to answer one without addressing one or more others as well. Many of these questions have been touched upon in publications, but none of them has a satisfying answer at this point (in my opinion). I provide some background and references for each question, and will try to lay out some potential pathways that could help to study them. Some of these questions are very tough and require (a group of) experts, while others are relatively straightforward and could, for example, make a nice master's project. Some questions require extensive and novel experiments while others could use rather standard

[2] See also: https://www.debtox.info/about_mechmod.html.

toxicity tests or even make some progress from existing data sets. I hope this book will inspire some of the readers to dive deeper into TKTD modelling: view it as an approach to learn what chemicals are doing in organisms, a strategy to test your hypotheses, rather than as 'just' another tool for risk assessment.

In the remainder of this book, the questions are grouped in chapters. Each chapter starts with a short introduction. Each section within each chapter deals with an open question (or topic of research), and references are provided at the end of each section (rather than at the end of the book). Each section is rather short (I aim for a maximum of two pages, including a figure and references), so each discussion is rather superficial. The references given provide more details (and more references to consider).

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Chapter 2

Toxicokinetics and damage dynamics

2.1 Introduction

Early TKTD models did not consider damage. Instead, the (predicted) internal concentration was directly linked to the effect mechanism (e.g., [2, 9]). However, we do need to consider damage for many compounds as it is clear that we cannot always explain the dynamics of effects from the kinetics of the chemical in the animal's body (for obvious examples, see [8, 1]). In the original publication of GUTS [5], damage was an optional 'dose metric', but with the first edition of the GUTS e-book in 2018 [6], damage received a central position: toxic effects are always related to damage, using either a full model (TK and damage dynamics as separate compartments) or a reduced model (TK and damage dynamics combined into a single compartment). This nicely consistent structure was later also adopted for DEBtox models [4, 7].

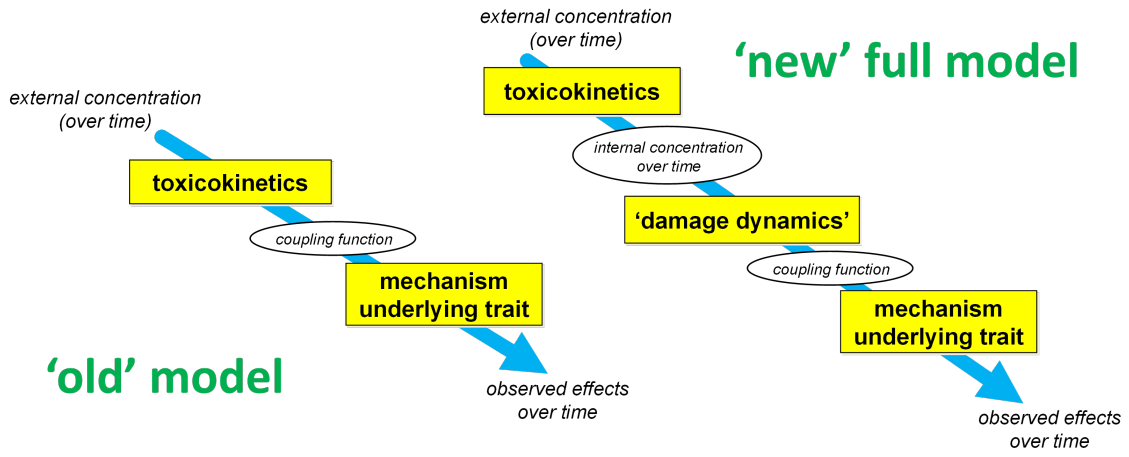

Figure 2.1: Early TKTD models did not include damage, while 'modern' models do.

The TK in TKTD is usually of the simplest form: the one-compartment model with first-order kinetics. However, the framework of TKTD modelling has no problem with more extensive TK modules. Damage is treated as an abstract one-compartment state variable with first-order dynamics. Damage is accrued from the internal concentration, and repaired with a certain rate constant. Since damage is not measured (and may not be measurable), scaling is used such that we only need to estimate a single rate constant for damage from the toxicity data (damage repair rate). The full model can be simplified to a reduced model,

by collapsing the two-compartment system into a one-compartment module (see Section 2.3).

For more details on TK and damage dynamics, I suggest the GUTS e-book ([6], Chapter 3). In more generality and detail, these subjects are treated in the e-book on mechanistic modelling ([3], Chapters 6 and 7).

This chapter deals with the following questions:	
What is damage?	Section 2.2
Does toxicokinetics or damage dominate?	Section 2.3
What feedback mechanisms are important?	Section 2.4
Do we need to consider biotransformation?	Section 2.5
Other exposure pathways?	Section 2.6

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2.2 What is damage?

Background. In the context of modern GUTS [5] and DEBtox [4, 6] models, damage is treated as an abstract one-compartment state variable with first-order dynamics. This is necessary as we want to keep things simple, and don't want these model frameworks to be chemical- and species-specific. Furthermore, we usually do not have (or do not want to use) information on the detailed mechanism of action of the chemical in the species of interest when analysing the results from toxicity tests. In general, this works fine. However, it would strengthen TKTD research to know more about the mechanisms, and this could help to identify cases where first-order damage dynamics is not sufficient.

Quite separate from the TKTD modellers, there are ecotoxicologists working on the molecular mechanisms of toxicity. Over the last decade, elucidating the chain of processes leading to toxicity is often referred to as adverse outcome pathways (AOP, see e.g., [1]). However, AOP has been notoriously sketchy when it comes to the link between sub-individual responses and organism response over time (e.g., growth, reproduction and survival). A closer collaboration between experts in TKTD and AOP would be beneficial in this respect.

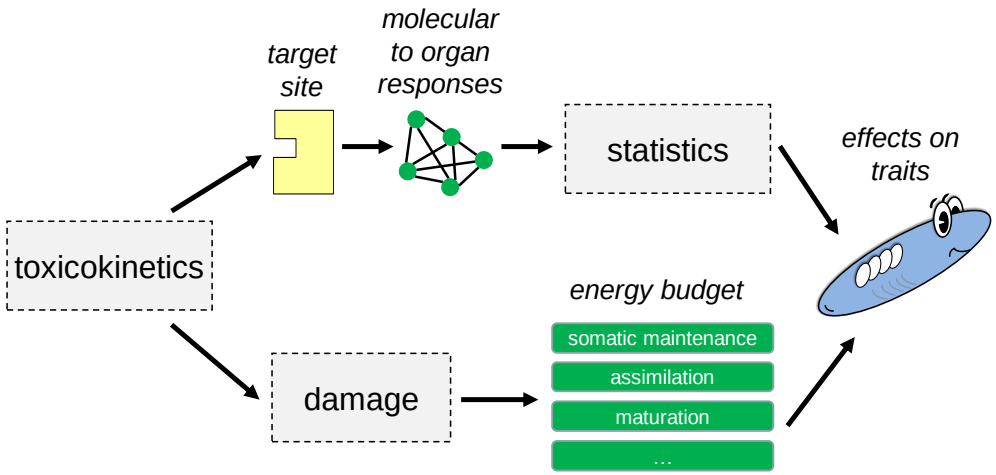

Figure 2.2: Schematic relationship between adverse-outcome-pathway-thinking (top chain) and DEBtox-thinking (bottom chain). Both attempt to link external concentrations via toxicokinetics to life-history traits of individuals. AOP is sketchy on the link between the molecular level and individual traits (and treats is as static), while DEBtox is sketchy on what 'damage' actually is. This figure from [2]

Possible approaches. Collaborate with ecotoxicologists that work on the mechanisms of action for specific (groups of) compounds (in specific species). A discussion and conceptual link between AOP and DEB was published by [8], for gene expression and GUTS see [9]. Some discussion was also presented in [3] and in the DEBtox e-book ([2], Section 5.4, which also contains many more useful references). I don't think that we should aim for extended AOPs (including DEB or GUTS models) to replace simpler TKTD approaches. AOPs will always be complex and difficult to parametrise, and will not be available for each chemical (group) and each species (group). However, a cross fertilisation has the potential to strengthen each field.

In this context, it is also useful to point at more specific, but still simple, models for damage. Particularly, I would like to draw attention to the receptor module of [7], where effects

are linked to the fraction of occupied receptors. A difference with the standard damage module is that the receptor module can saturate at high concentrations (when all receptors are occupied, the effects cannot get worse).

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2.3 Does toxicokinetics or damage dominate?

Background. The recent (since 2018) implementations of GUTS and DEBtox apply the distinction between 'full' and 'reduced' models. In full models, there are two compartments driving the toxic effects: toxicokinetics (i.e., internal concentration) and damage dynamics. The internal concentrations leads to production of (scaled) damage. In reduced models, these two are collapsed into a single scaled damage compartment by assuming that one of these processes dominates the overall dynamics. In the absence of measured internal concentrations, reduced models are typically used as these can often be parametrised from effects over time only. Although, in exceptional cases, we can use a full model (with scaled TK) without measured body residues [2, 4].

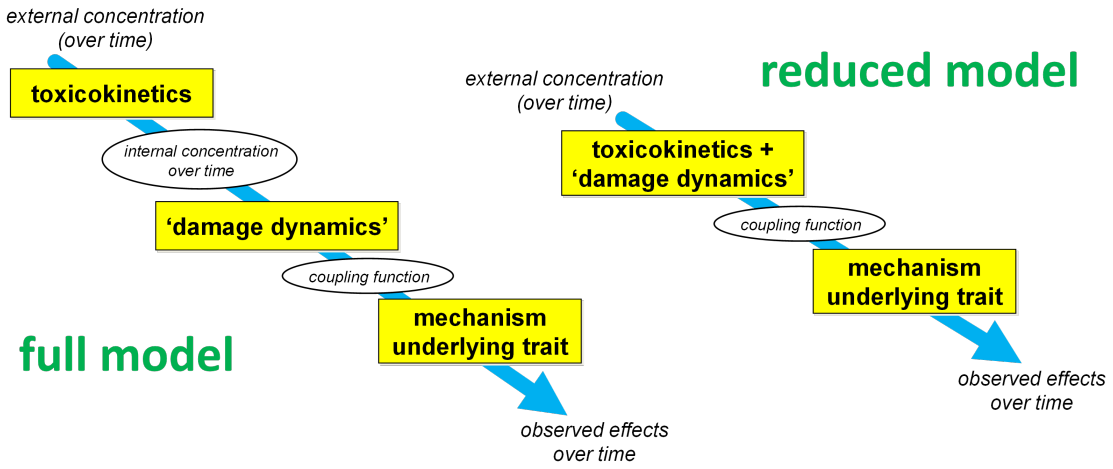

Figure 2.3: The distinction between full and reduced TKTD models.

When using a reduced model, does it matter whether TK or damage dynamics dominates? For some questions, it does. When we estimate the dominant rate constant from toxicity data, we cannot know *a priori* whether it is a TK elimination rate or a damage repair rate. This also implies that we should not use reported TK elimination rates (or QSAR predictions) as input for TKTD models to predict effects, unless we can be sure that damage dynamics is very fast. A second issue arises when animals grow and/or reproduce in the toxicity test, which will affect TK but may not affect damage dynamics. This situation is especially common with DEBtox applications and is treated in Section 2.4. Furthermore, it is good to know when TK and damage dynamics are of a similar speed (and rather slow). That would imply that we may need to consider using a full model (two-compartment damage dynamics) rather than a reduced one (one-compartment damage dynamics).

Possible approaches. If we only have toxicity data, there is generally no way of knowing whether TK or damage dynamics has dominated the overall effect dynamics. If we have knowledge on the chemical's mechanism of action, in the species of interest, we may be able to make an educated guess. For narcotic compounds, we may expect damage dynamics to be very fast, while for organophosphates, we may expect it to be very slow. If we have measured body residues over time, we can use a full model and quantify the speed of damage dynamics explicitly [1], and compare it to the TK elimination rate. We can also look at chemicals with a known mode of action and see whether the dominant rate constant, as

derived from the toxicity data, aligns with what we can expect for the TK elimination rate [3]. It is important to note that the last two references are with survival models (GUTS and a predecessor). An added complication for survival is the uncertainty about whether SD or IT is the dominant mechanism for mortality. The selection of the death mechanism affects the values of the rate constants [1]. Therefore, this question is best tackled in conjunction with the question on the most appropriate death mechanism (see Section 3.3), and perhaps also considering biotransformation (see Section 2.5).

The answer to whether TK or damage dynamics dominates depends on the species and on the chemical, and perhaps on more factors (e.g., body size, temperature). Therefore, it is unlikely that the answer is a simple one. Nevertheless, it would be good to have a better understanding of the speed of the processes that lead to damage production.

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