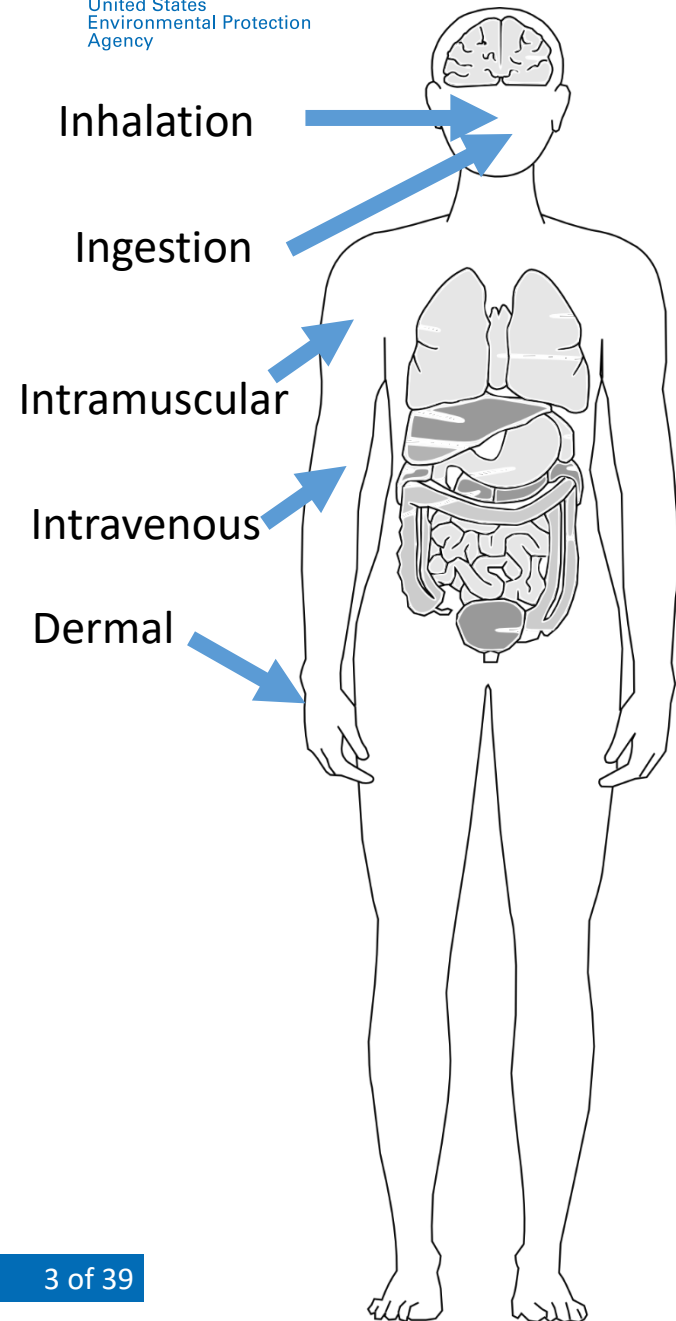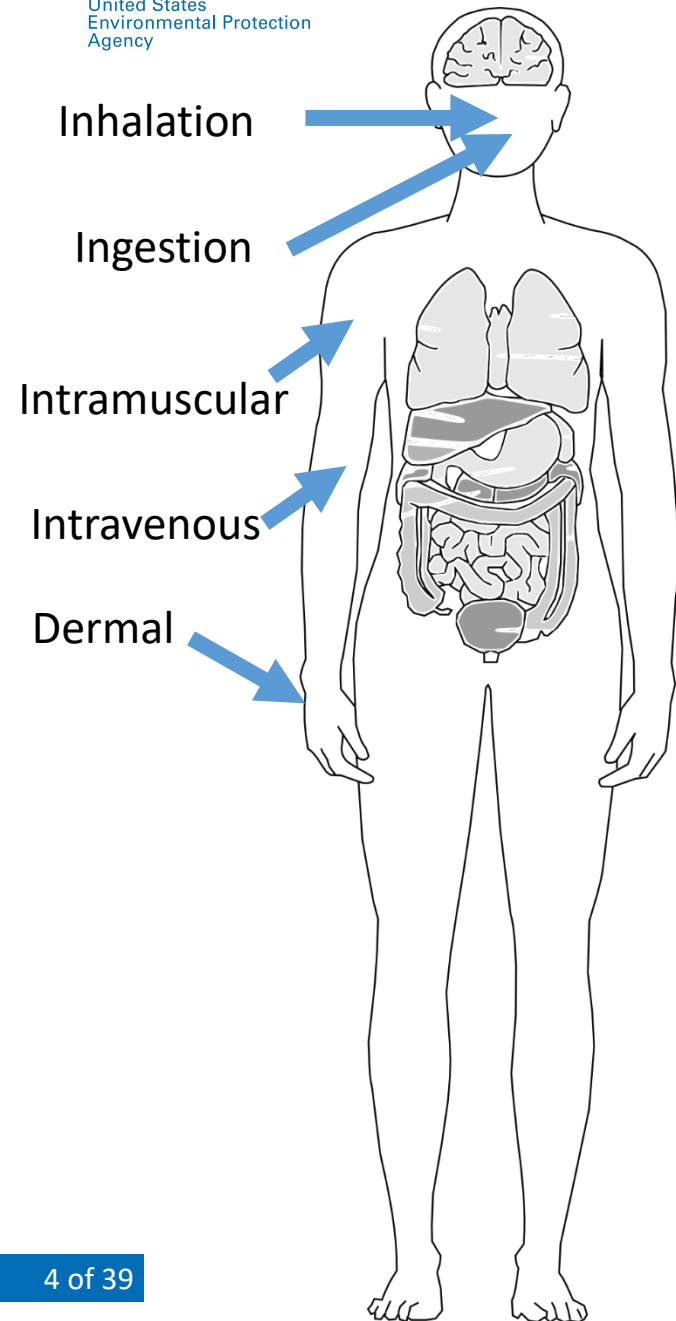# Toxicokinetics

Caroline L. Ring, Ph.D

Overview

- Motivating scenarios: How does the body process a chemical?
- What is toxicokinetics (TK)?
- How does toxicokinetics describe how the body processes a chemical?
- One-compartment model
- Two-compartment model
- Summary metrics of internal exposure
- Physiologically-based TK models
- Applications of TK models
  - Inter-species extrapolation
  - Internal-external extrapolation
  - Route-to-route extrapolation
  - In vitro-in vivo extrapolation (IVIVE)
- Activity: Carrier-Aylward model

# Scenario: You are exposed to a chemical

Inhalation

Ingestion

Intramuscular

Intravenous

Dermal

Inhalation

Ingestion

Intramuscular

Intravenous

Dermal

Scenario: You are exposed to a chemical

Things you might want to know….

- Does the chemical get inside your body?

- If so, how much of it gets inside? For example, what is the concentration of chemical in your blood?

- Is that enough to cause any kind of health effect? (desired or undesired)

- Does it settle in any particular tissues or organs?

- Does it stay in your body for a long time, or does your body excrete it quickly?

Another scenario: You are a regulator who needs to set a limit on *external* exposure to Chemical X, but you only have data that relates negative health effects with *internal* blood levels of Chemical X.


Figure adapted from Mocarelli et al. (2008)
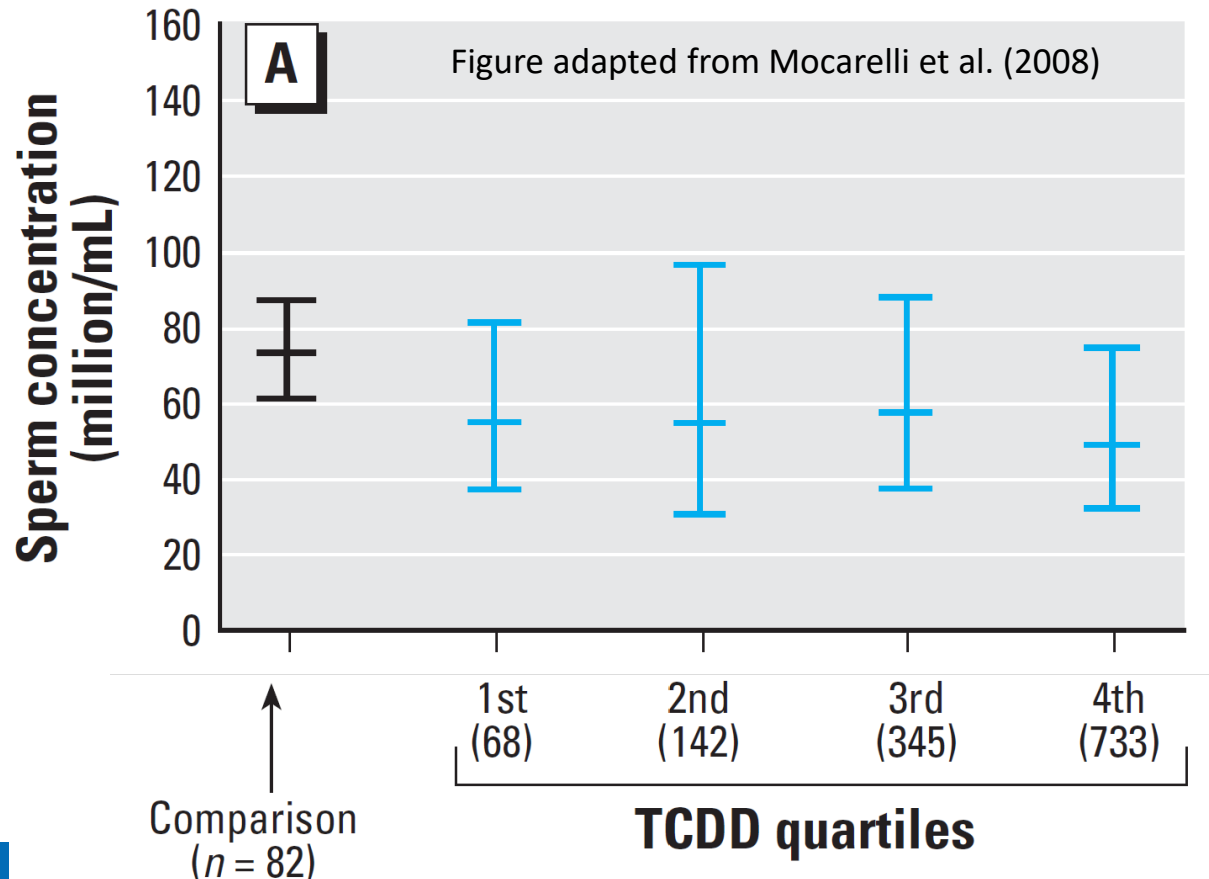
Example: In 2012, the US EPA published[1] a reference dose (external exposure) for 2,3,7,8-TCDD.

It was based in part on a study[2] of sperm concentration in men who had been exposed to high levels of TCDD as children, after a major industrial accident in Seveso, Italy.

The EPA determined that, in this study, sperm concentration was reduced when TCDD concentration in blood serum (adjusted for lipid content) was 68 ppt.

What external exposure to TCDD would produce that blood level?

[1]US EPA (2012), EPA/600/R-10/038F

[2]Mocarelli et al. (2008), DOI 10.1289/ehp.10399
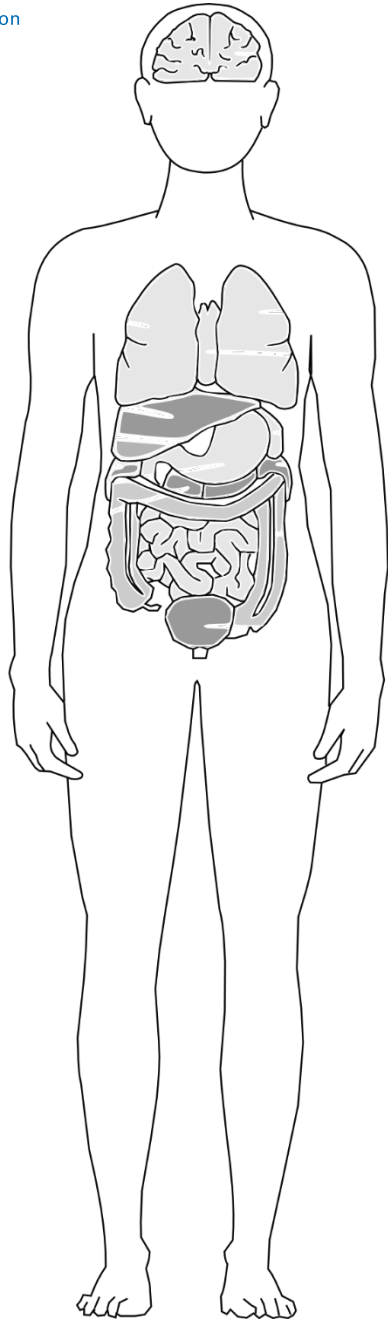
Toxicokinetics (TK) answers these questions by describing "what the body does to the chemical"

(as opposed to *toxicodynamics* [TD], which describes "what the chemical does to the body")

ADME

- **Absorption**: How does the chemical get absorbed into the body tissues?

- **Distribution**: Where does the chemical go inside the body?

- **Metabolism**: How do enzymes in the body break apart the chemical molecules?

- **Excretion**: How does the chemical leave the body?

Pharmacokinetics (PK) is a synonym for toxicokinetics (TK). They are often used interchangeably. PK connotes pharmaceuticals; TK connotes environmental chemicals – but those connotations are weak.

TK does *not* describe any effects the chemical might have on the body (toxic, therapeutic, or other)
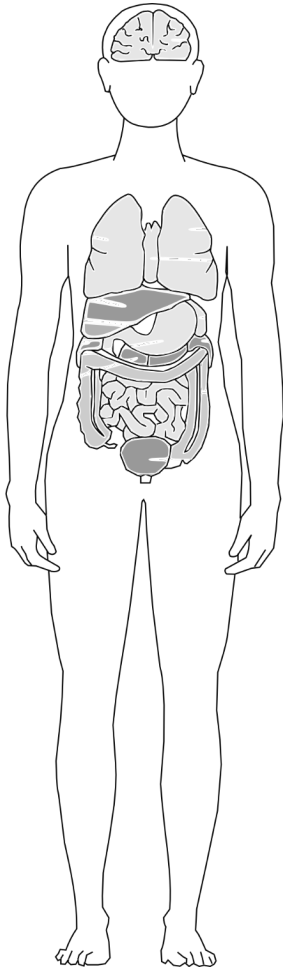
TK tells you where the chemical goes, how fast it goes there, and how much of it goes there –
but not what it does when it gets there!

The only exception is that sometimes, "what the chemical does" includes some effect on ADME.

For example, for some chemicals, your body might sense a high concentration and send a signal to your liver to boost metabolism of that chemical. A TK model might describe that kind of feedback effect.
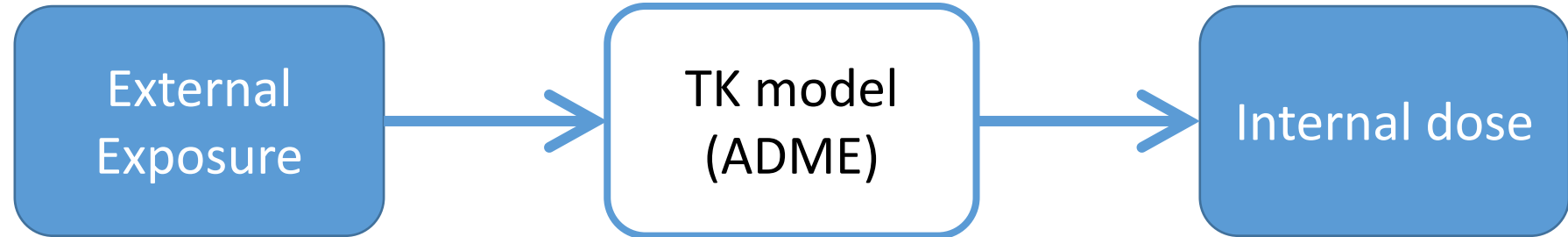
But TK models are very useful in combination with data or models about toxicity – as we'll see later!

Toxicokinetics links external exposures to internal doses

External exposure
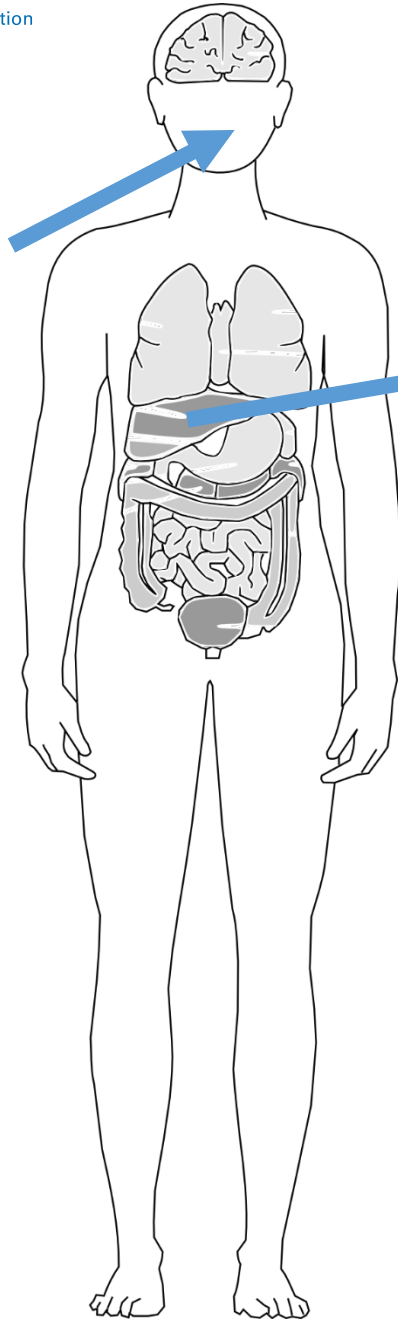
**Internal dose** = Concentration of chemical or drug in one or more body tissues of interest

External Exposure	→	TK model (ADME)	→	Internal dose

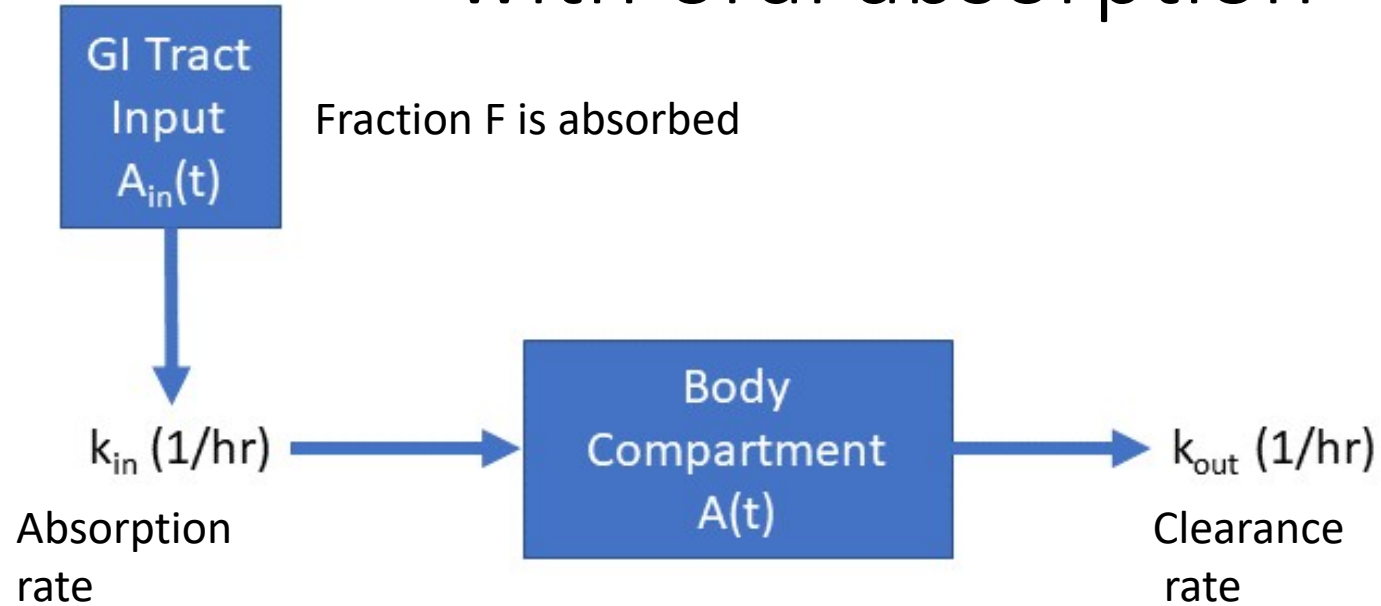# TK models describe ADME mathematically by representing the body as compartments and flows
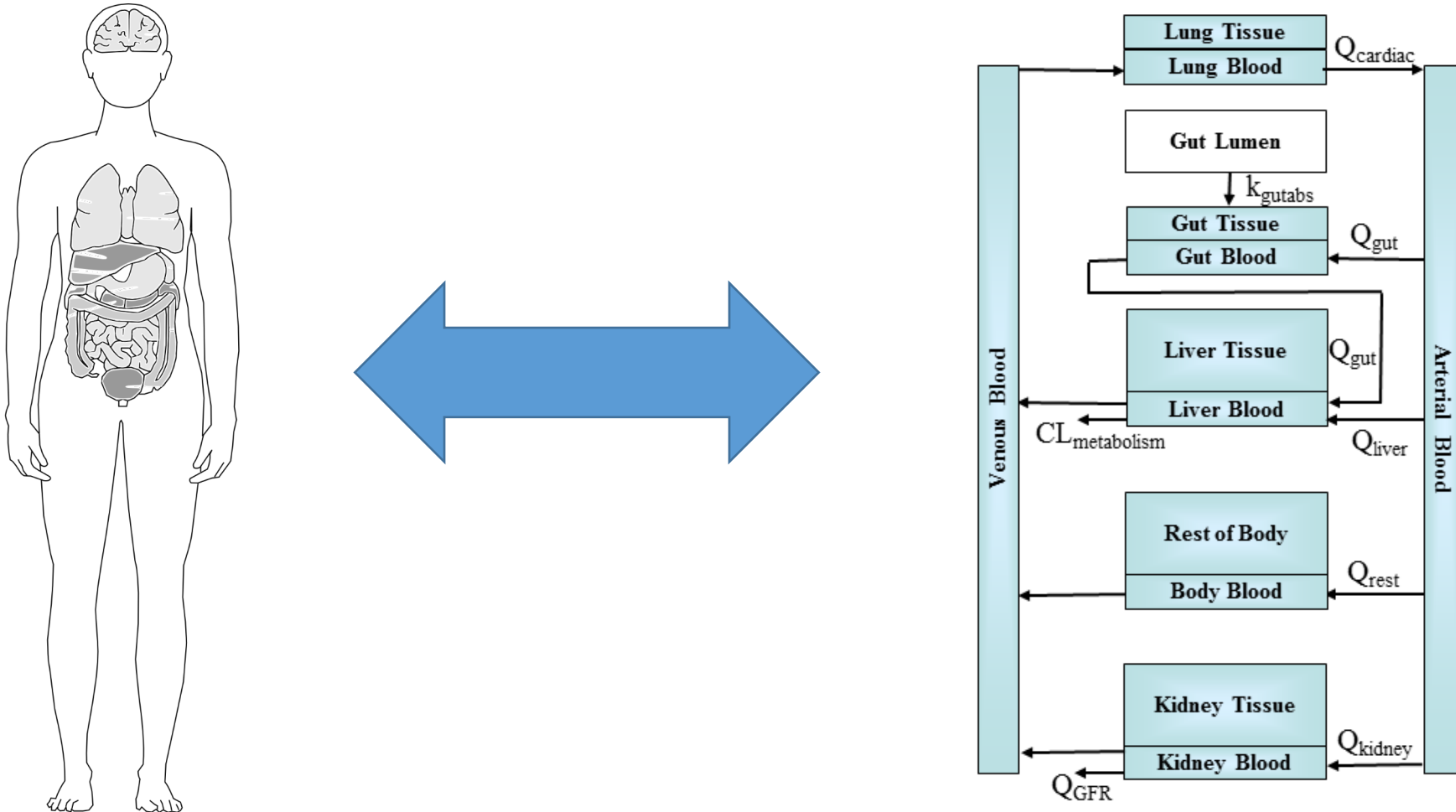
The amount of chemical in each compartment (vs. time) is described using a mass balance equation

Rate of change of amount in compartment k =
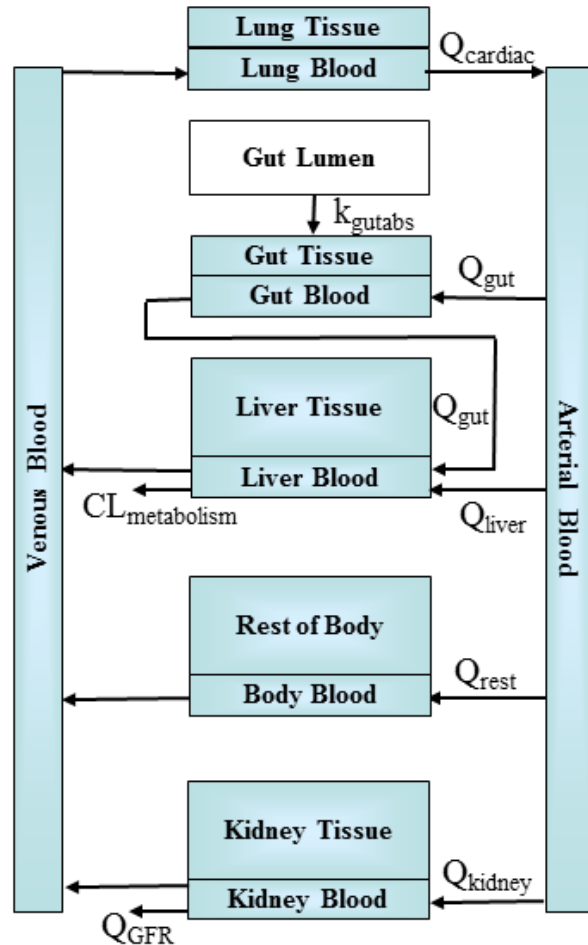
$$\frac{dA_k}{dt} =$$

[Amount entering compartment k per unit time] – [Amount leaving compartment k per unit time]

A simple TK model: one compartment with oral absorption

GI Tract Input $A_{in}(t)$

Fraction F is absorbed

k_{in} (1/hr)
Absorption rate

Body Compartment $A(t)$

k_{out} (1/hr)
Clearance rate

Assume body is one big well-mixed compartment

Bourne, D. (2021).
https://www.boomer.org/c/p4/

A simple TK model: one compartment with oral absorption

Fraction F is absorbed

k_{in} (1/hr)

Absorption rate

k_{out} (1/hr)

Clearance rate

Assume body is one big well-mixed compartment

Mass balance equations:

$$\frac{dA}{dt} = k_{in}FA_{in}(t) - k_{out}A(t)$$

$$\frac{dA_{in}}{dt} = -k_{in}FA_{in}(t)$$

Bourne, D. (2021). https://www.boomer.org/c/p4/

A simple TK model: one compartment with oral absorption

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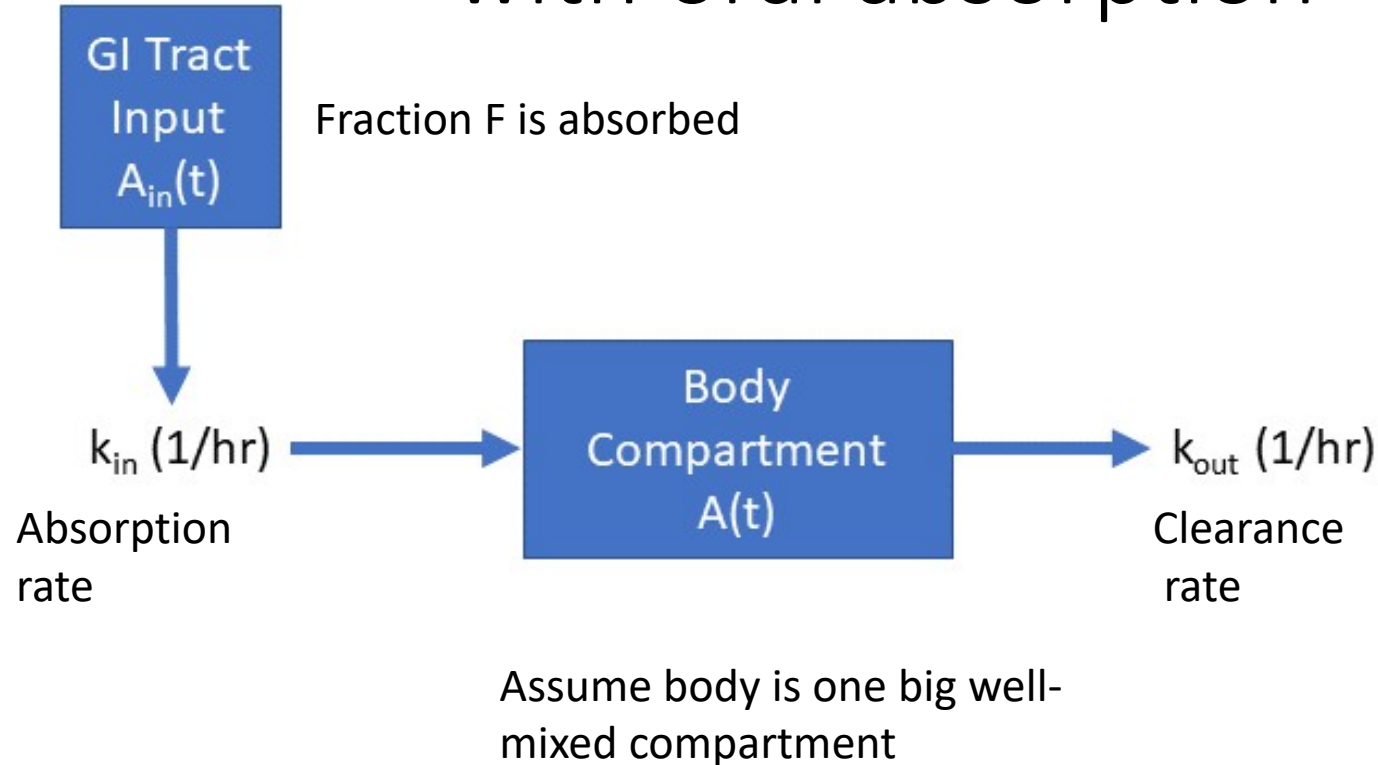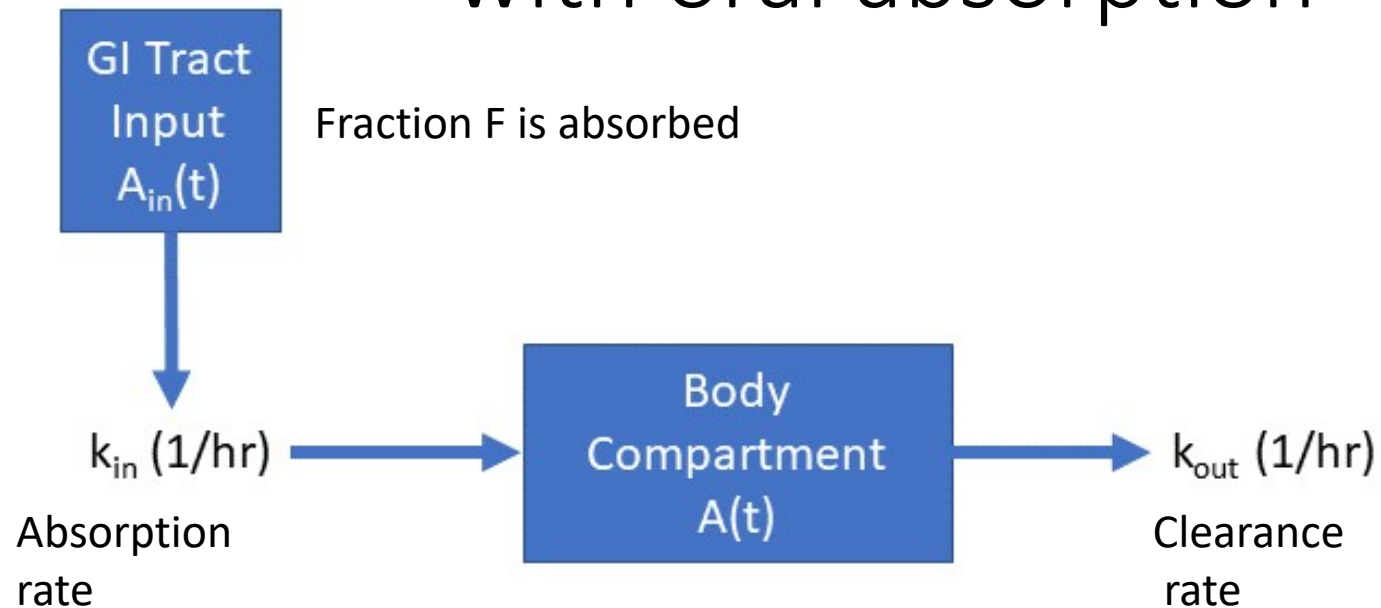**Mass balance equations:**

$$\frac{dA}{dt} = k_{in}FA_{in}(t) - k_{out}A(t)$$

$$\frac{dA_{in}}{dt} = -k_{in}FA_{in}(t)$$

Initial conditions:

$$A_{in}(t = 0) = \text{administered amount}$$
$$A(t = 0) = 0$$

Bourne, D. (2021).
https://www.boomer.org/c/p4/

A simple TK model: one compartment with oral absorption

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Initial conditions:

$$A_{in}(t = 0) = \text{administered amount}$$
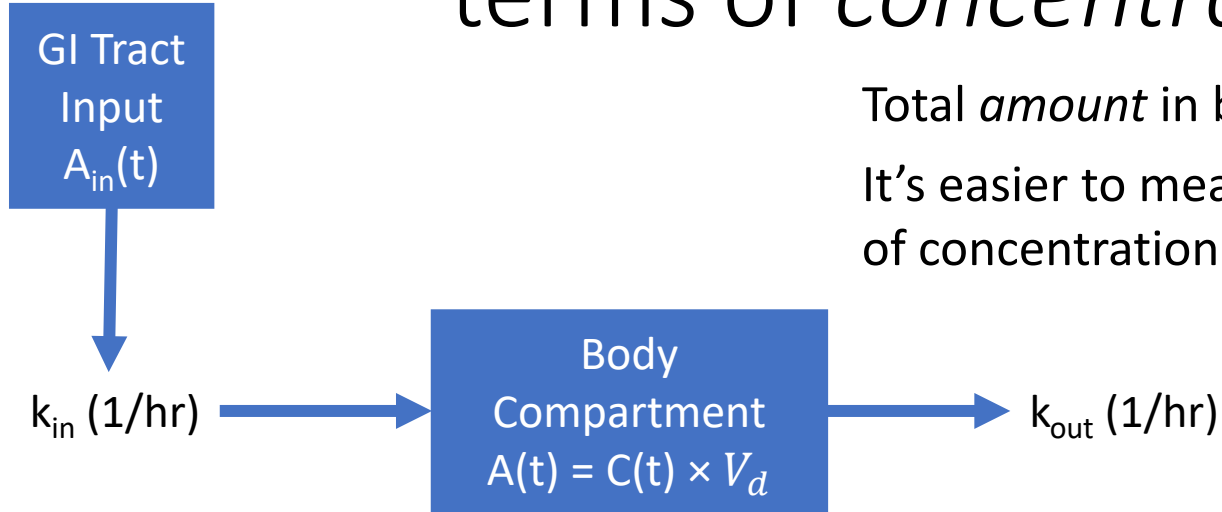$$A(t = 0) = 0$$

Analytical solution:

$$A(t) = \frac{F\, A_{in}(0)\, k_{in}}{k_{in} - k_{out}} \left[e^{-k_{out}\, t} - e^{-k_{in}\, t} \right]$$

Bourne, D. (2021).
https://www.boomer.org/c/p4/

One-compartment model solution in terms of *concentration* vs. time

GI Tract Input $A_{in}(t)$

k_{in} (1/hr)

Body Compartment $A(t) = C(t) \times V_d$
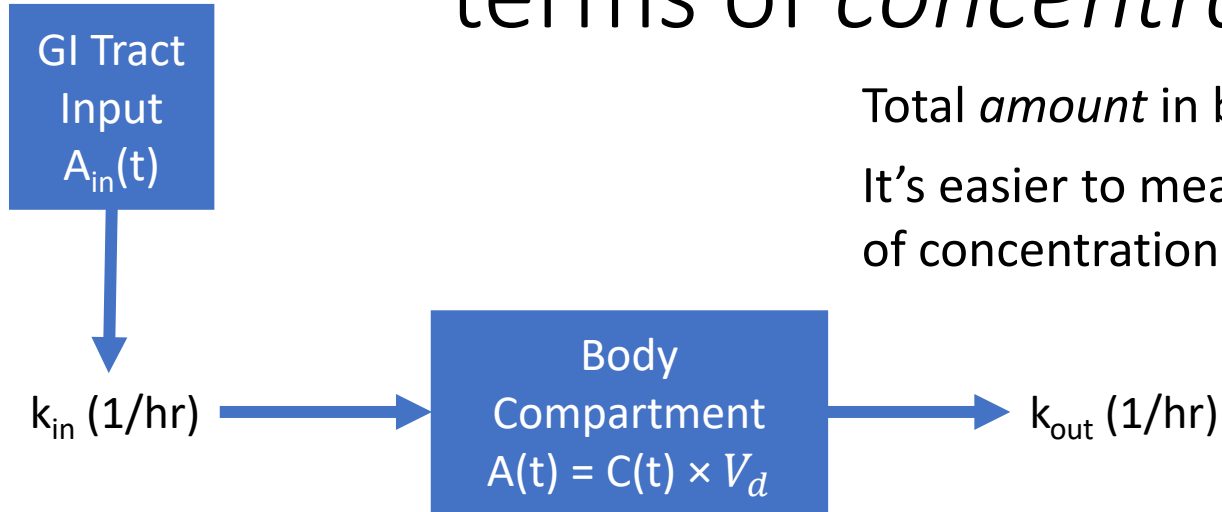
k_{out} (1/hr)

Total *amount* in body (body burden) is difficult to measure

It's easier to measure *concentration* – so re-express in terms of concentration

$$C(t) = \frac{F\, A_{\text{in}}(0)\, k_{\text{in}}}{V_d(k_{\text{in}} - k_{\text{out}})} \left[e^{-k_{out}\, t} - e^{-k_{in}\, t} \right]$$

Amount in body [mg] = Concentration in body $\left[\dfrac{\text{mg}}{\text{L}}\right] \times$ Volume of distribution V_d [L]

Bourne, D. (2021). https://www.boomer.org/c/p4/

One-compartment model solution in terms of *concentration* vs. time

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Amount in body [mg] = Concentration in body $\left[\dfrac{\text{mg}}{\text{L}}\right] \times$ Volume of distribution V_d [L]

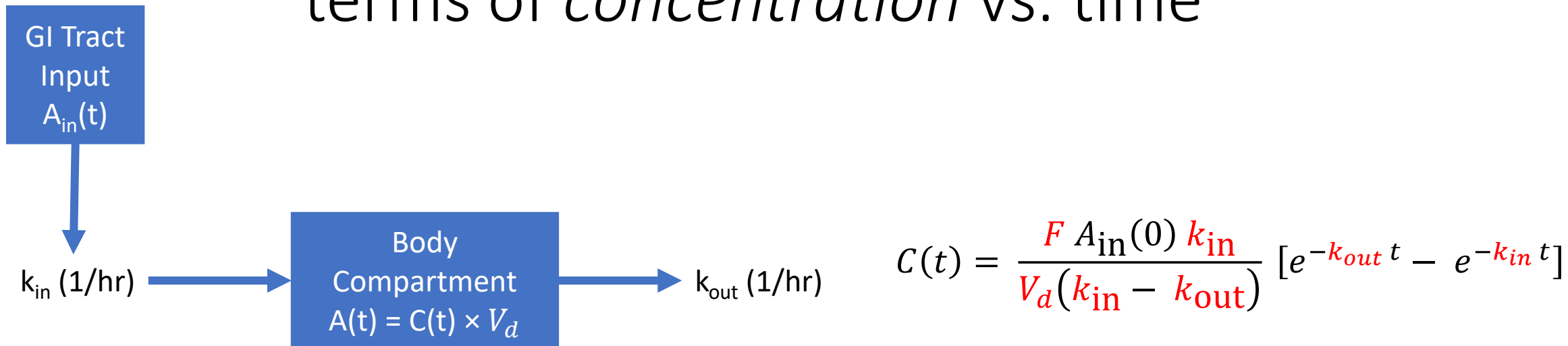V_d **does not represent the actual, physical volume of the body.**

V_d is a theoretical quantity.

If we put a known amount A directly into the body (e.g. IV), and then measure a concentration C in the blood, what volume V_d *would* be needed to make A and C obey A = C × V_d ?

(If V_d is very different from the physical body volume, it probably means the well-mixed one-compartment model is not a very accurate assumption!)

Bourne, D. (2021). https://www.boomer.org/c/p4/

One-compartment model solution in terms of *concentration* vs. time

GI Tract Input $A_{in}(t)$

k_{in} (1/hr)

Body Compartment $A(t) = C(t) \times V_d$

k_{out} (1/hr)

$$C(t) = \frac{F\, A_{in}(0)\, k_{in}}{V_d(k_{in} - k_{out})} [e^{-k_{out}\, t} - e^{-k_{in}\, t}]$$

For a given chemical –
how could we estimate one-compartment TK model parameters
F, V_d, k_{in}, k_{out} ?

Bourne, D. (2021).
https://www.boomer.org/c/p4/

TK parameters can be estimated by measuring body concentration vs. time for a known dose

Administer a known dose (mg/kg body weight)
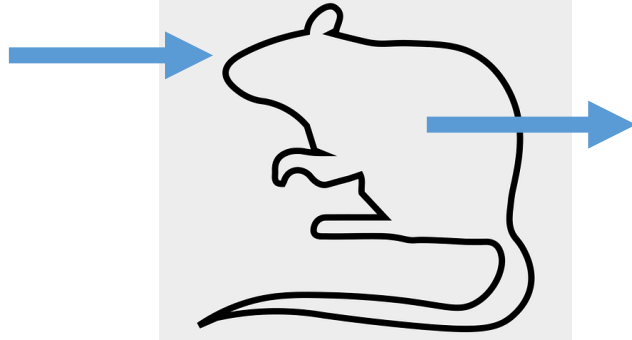
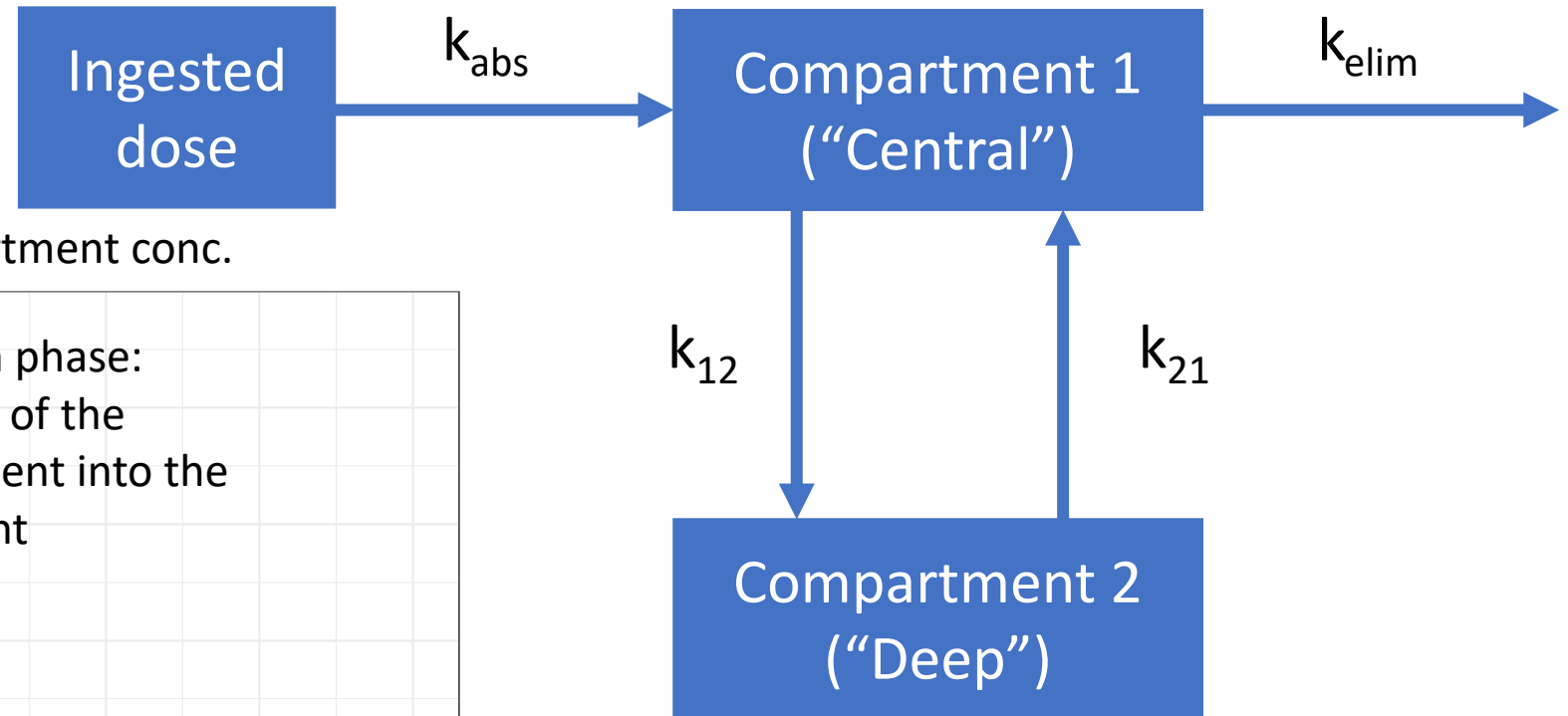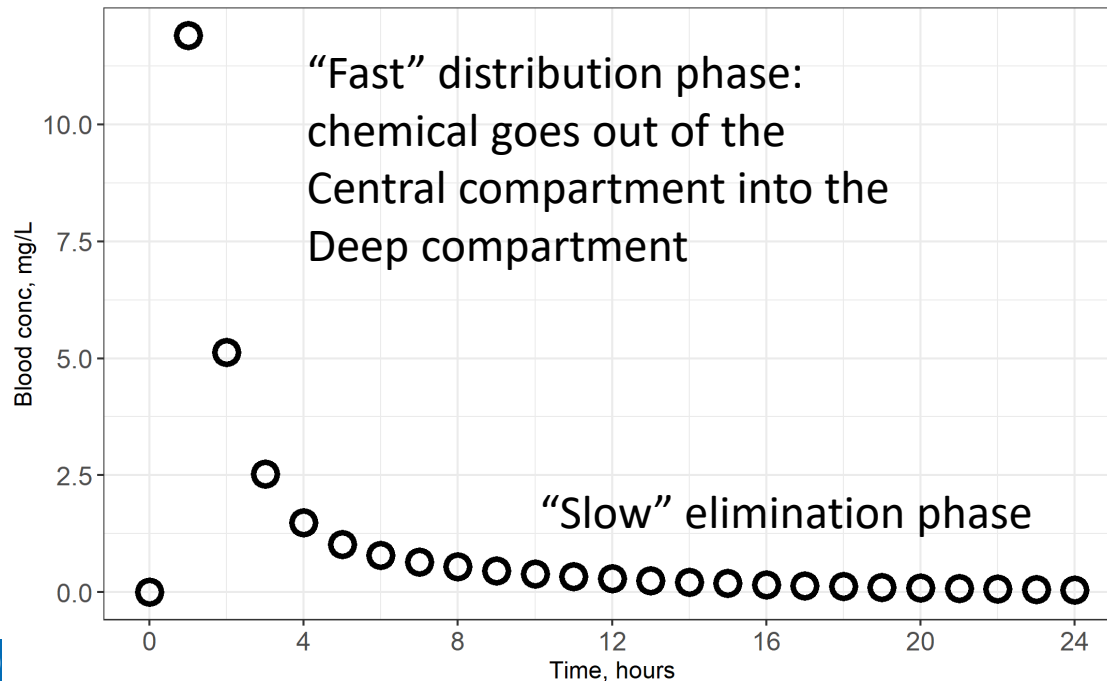Measure blood concentration at several time points

Fit 1-compartment model equation to concentration vs. time data

$$C(t) = \frac{F\,A_{\text{in}}(0)\,k_{\text{in}}}{V_d(k_{\text{in}} - k_{\text{out}})}\left[e^{-k_{out}\,t} - e^{-k_{in}\,t}\right]$$

Blood conc, mg/L

Time, hours
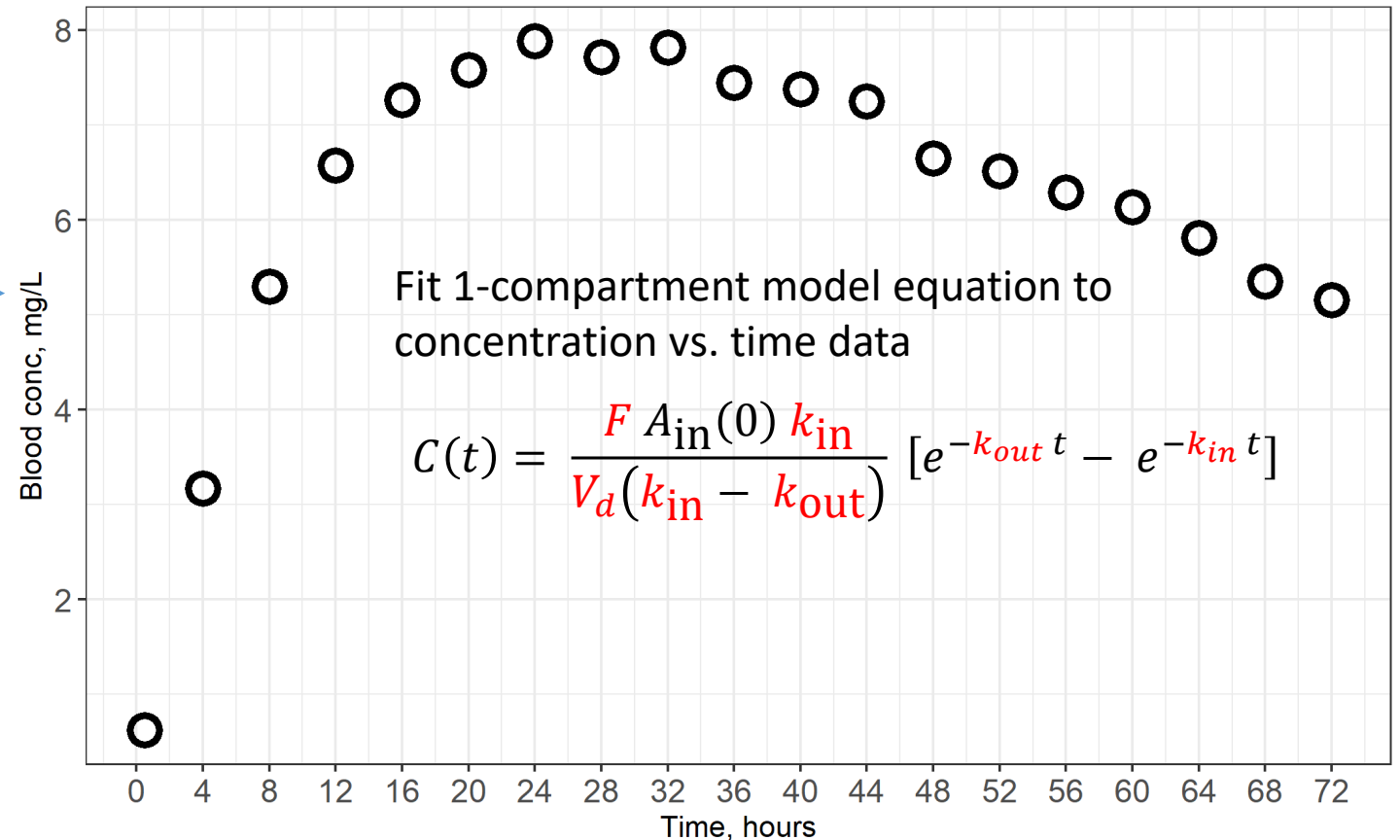
Lots more details can be found at Bourne, D. (2021). https://www.boomer.org/c/p4/

Concentration vs. time data may empirically be better described by a model with multiple compartments, e.g. two compartments

Assume blood conc. = Central compartment conc.

"Fast" distribution phase: chemical goes out of the Central compartment into the Deep compartment

"Slow" elimination phase

$$\text{Ingested dose} \xrightarrow{k_{abs}} \text{Compartment 1 ("Central")} \xrightarrow{k_{elim}}$$

k_{12} k_{21}

Compartment 2 ("Deep")

"Central" and "Deep" are again *theoretical* compartments – they don't represent actual, physical organs/tissues

Bourne, D. (2021). https://www.boomer.org/c/p4/

TK models can be used to calculate various time-independent summary metrics of internal dose

C_{max}:
maximum or "peak" concentration in the body/tissue

t_{peak}= time of peak concentration

Blood conc, mg/L

Time, hours

TK models can be used to calculate various time-independent summary metrics of internal dose

AUC:
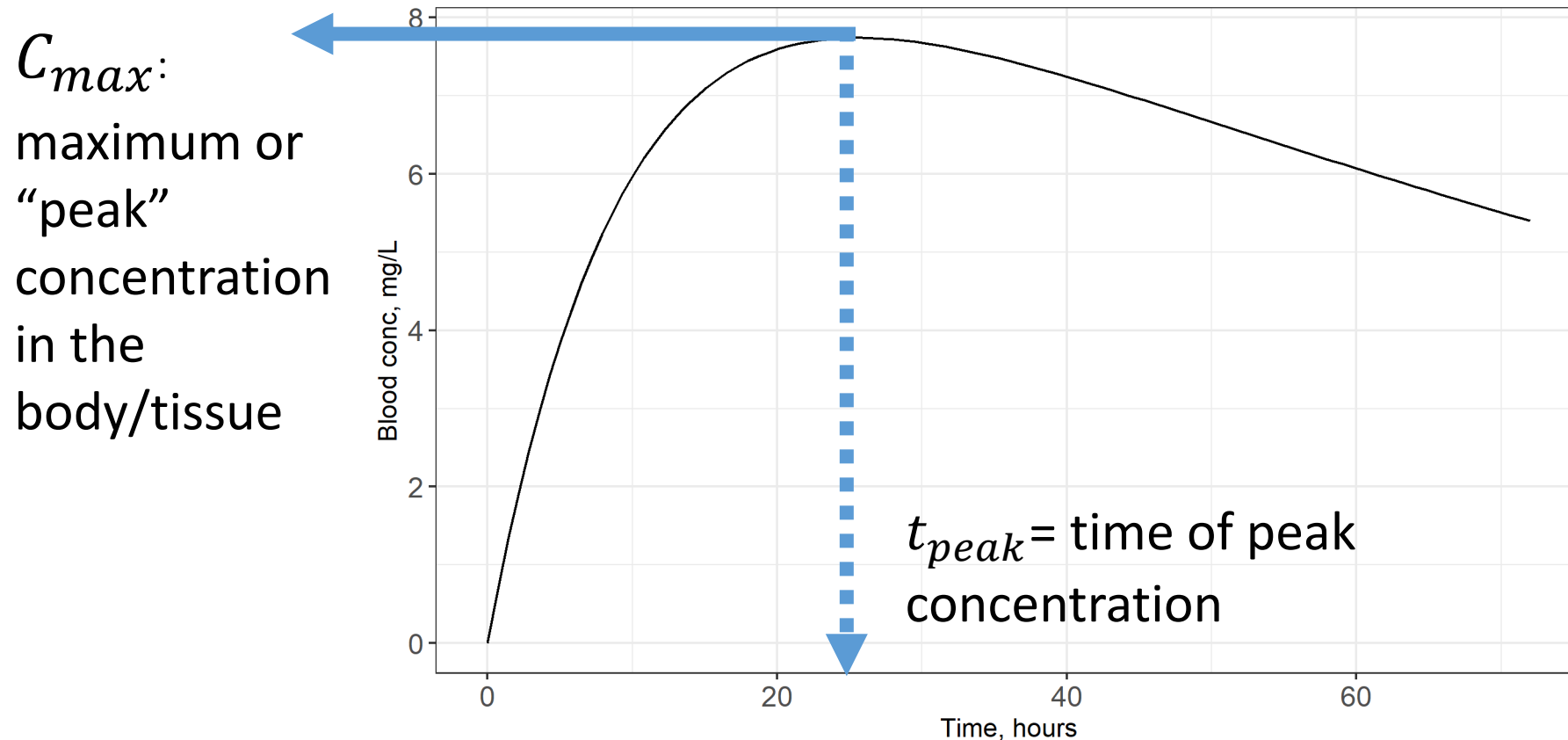
area under concentration/time curve = "cumulative dose"
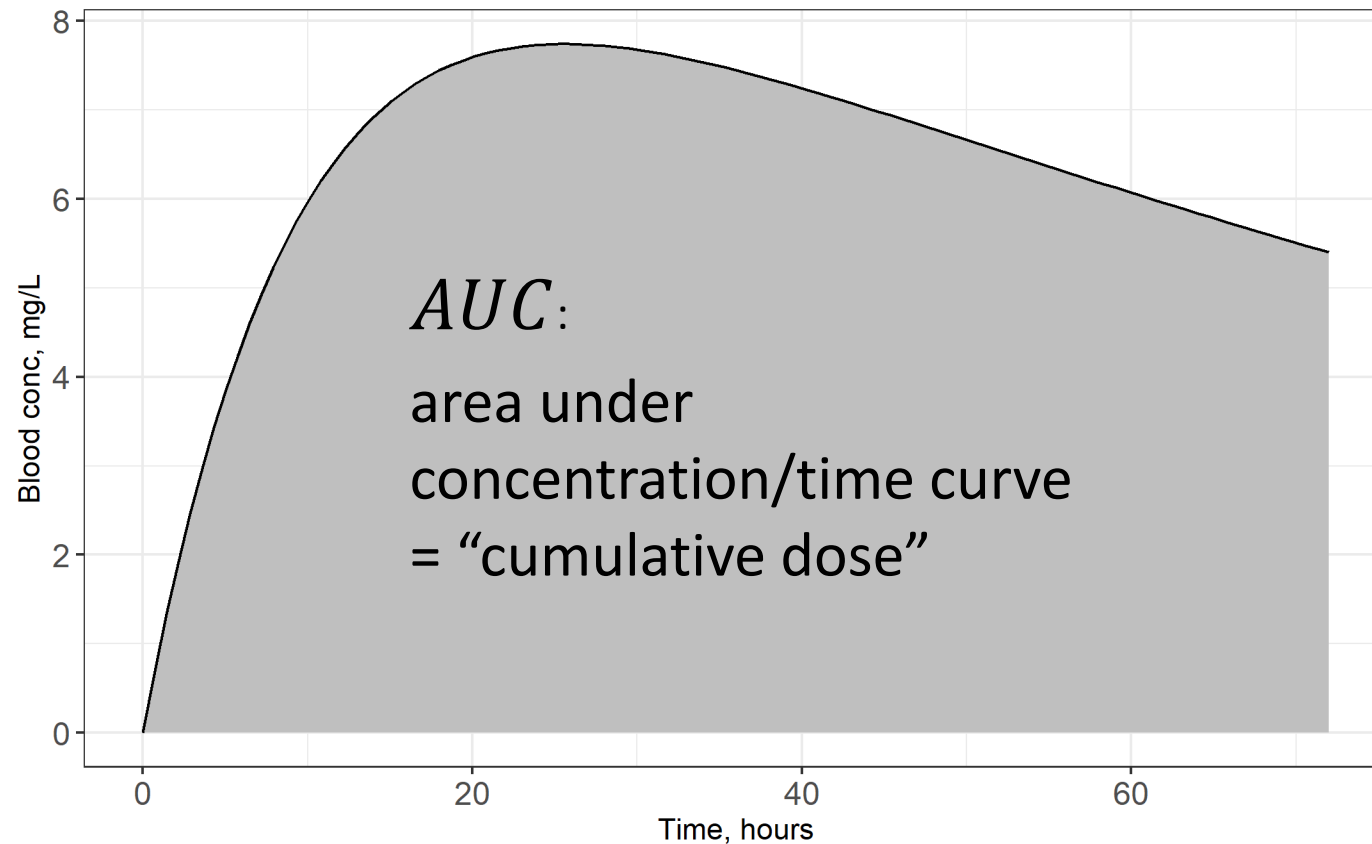
TK models can be used to calculate various time-independent summary metrics of internal dose
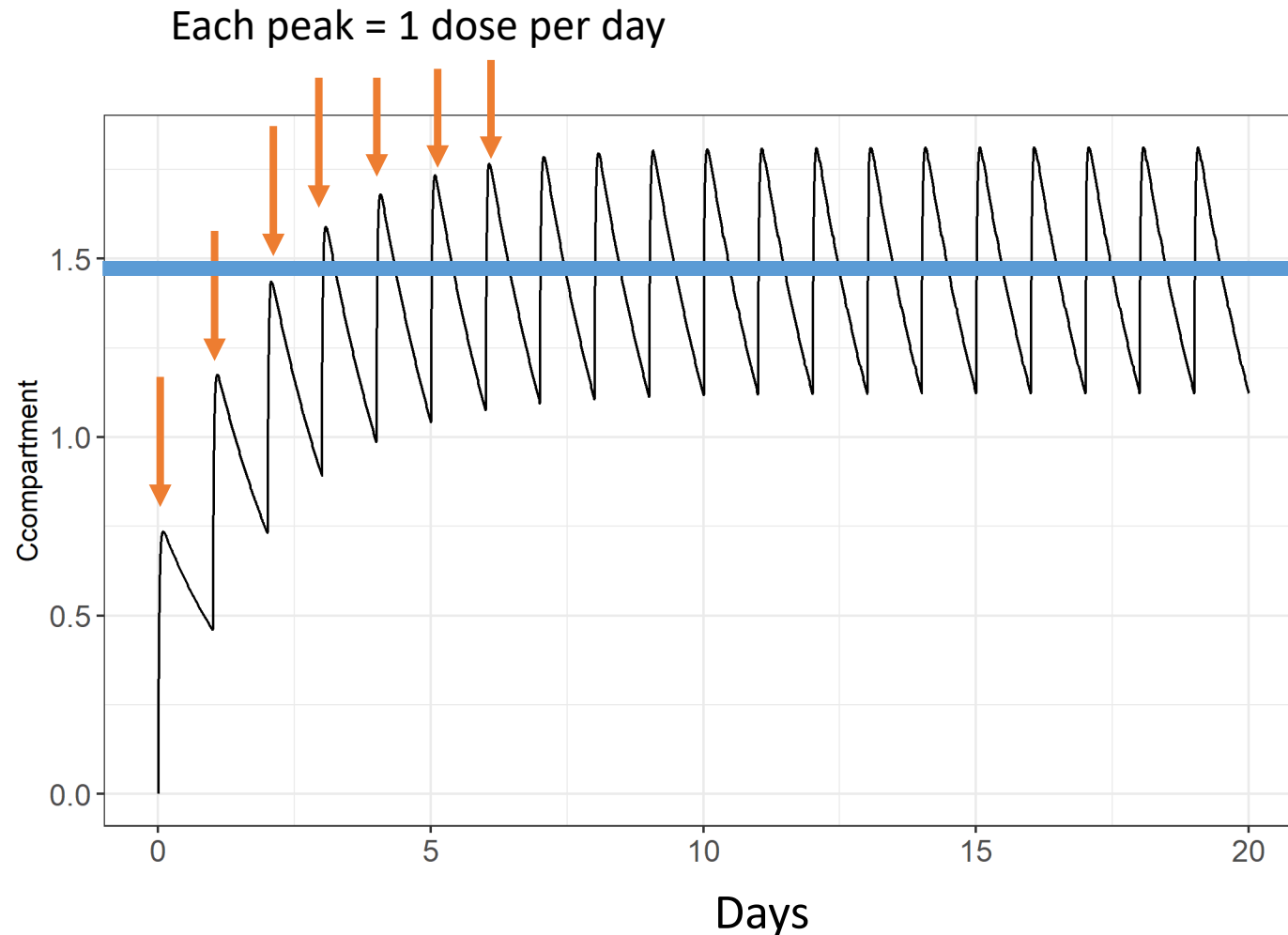
$$C_{mean} = \frac{AUC}{\text{total time}}$$

Average body/tissue concentration over time
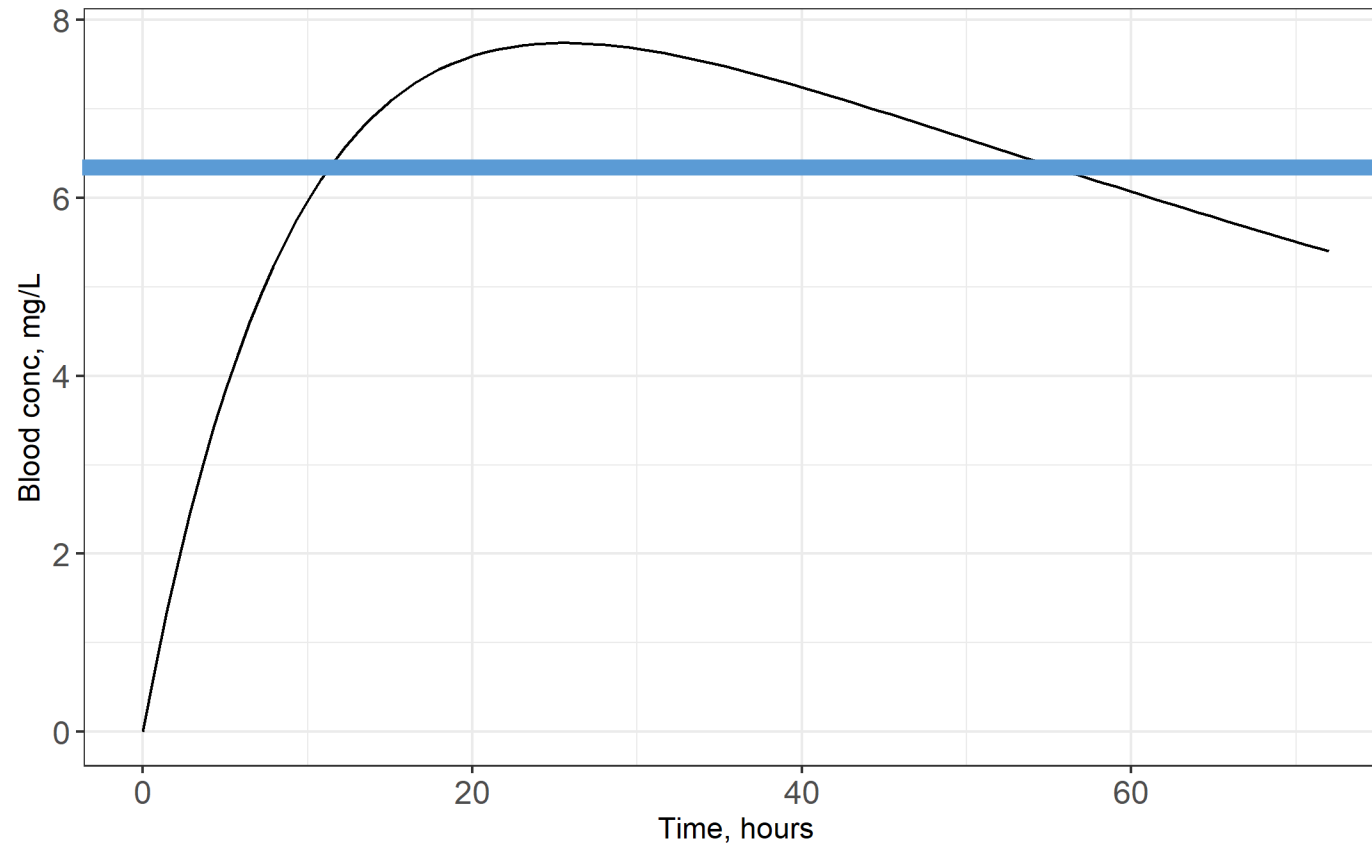
TK models can be used to calculate various time-independent summary metrics of internal dose

C_{ss}

Steady-state average body/tissue concentration after repeated dosing for a long time

Each peak = 1 dose per day

Ccompartment

Days

# Empirical compartmental models have benefits and drawbacks

Benefits

- Parsimonious: Lets you find the simplest model that fits concentration vs. time data

- Computationally simpler: Simple models have analytical solutions, meaning you don't have to integrate numerically

Drawbacks

- Data-intensive: Requires *in vivo* concentration vs. time measurements to fit for each new chemical, new species, new formulation…

- Interpretation: Parameters don't have direct physiological interpretation (like Vd), so it's harder to draw concrete physiological conclusions

Breen et al. (2021)

Rather than empirical 1- and 2- compartment models, we can use physiologically-based TK (PBTK) models, where compartments and flows now represent real physiological quantities

Empirical 1-compartment model

Empirical 2-compartment model
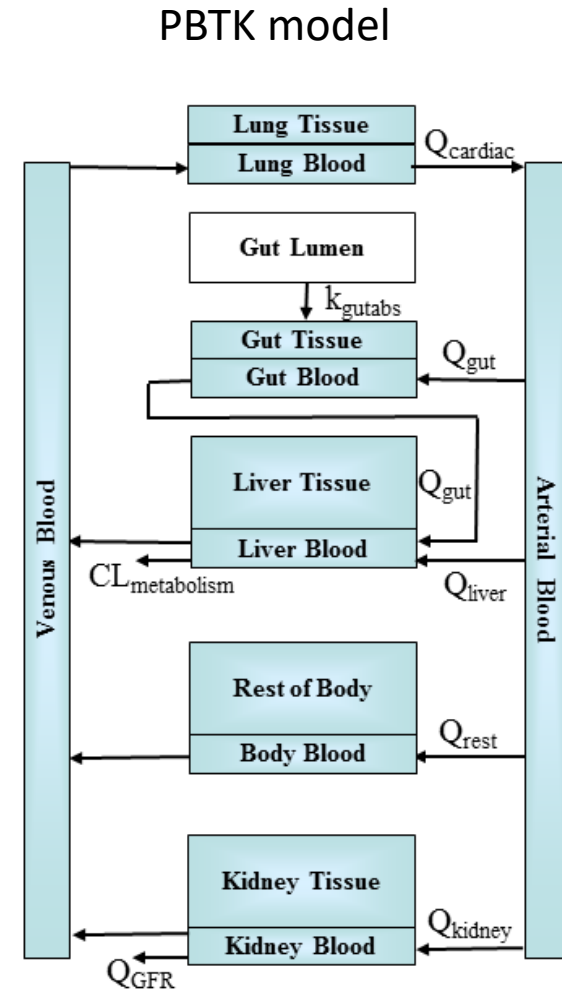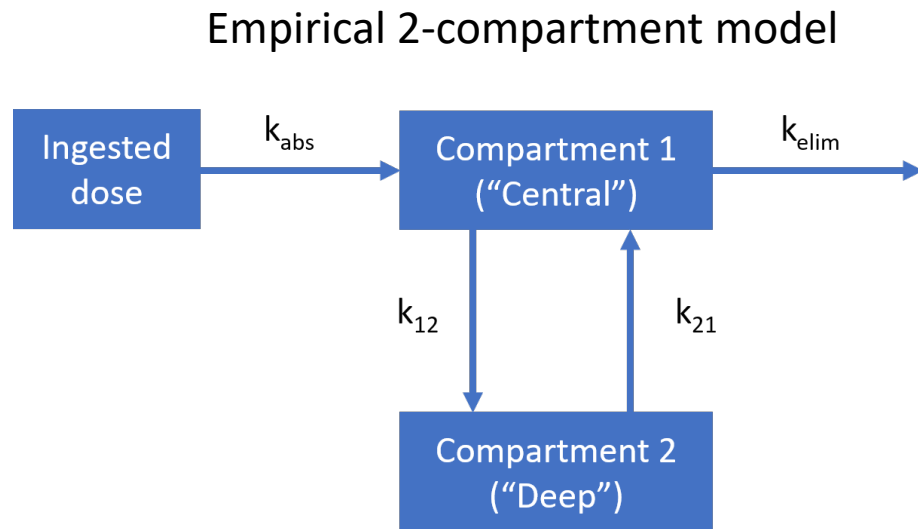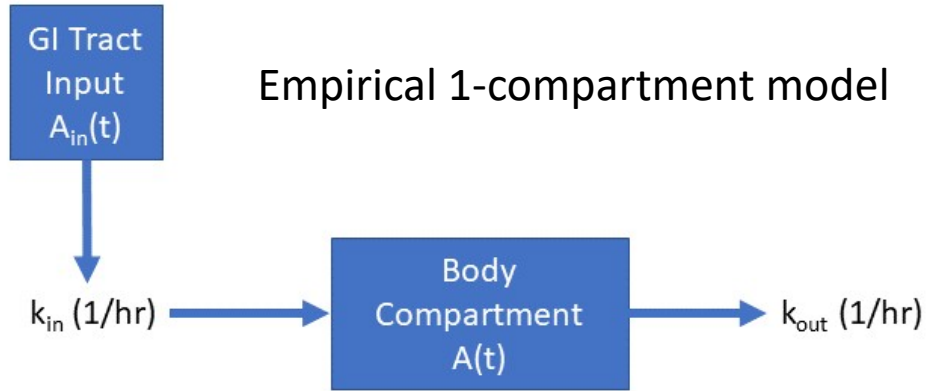
PBTK model

PBTK models have more parameters overall, but most of them don't need to be estimated from concentration vs. time data.

With empirical 1- or 2-compartment models, the *only* way to estimate model parameters is by gathering and fitting concentration vs. time data.

Example: Volume of distribution ≠ actual body volume!
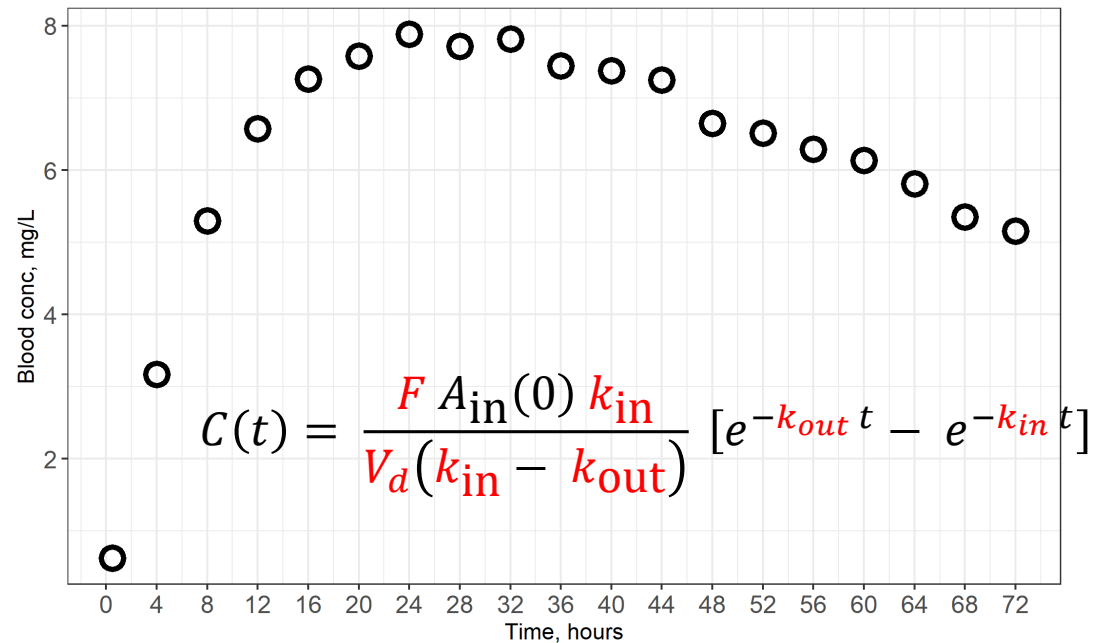
With PBTK models, parameters representing physiological quantities are known *a priori* based on studies of anatomy.

Example: Volume of liver compartment = actual volume of liver.

$$C(t) = \frac{F\, A_{in}(0)\, k_{in}}{V_d\left(k_{in} - k_{out}\right)} \left[e^{-k_{out}\, t} - e^{-k_{in}\, t}\right]$$
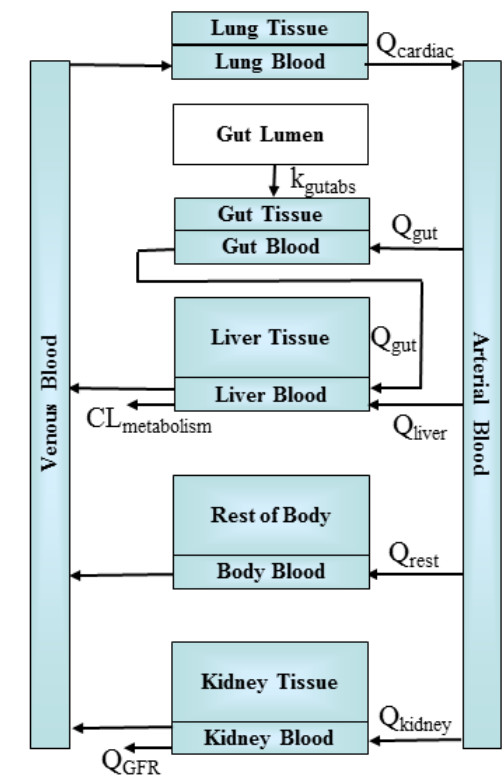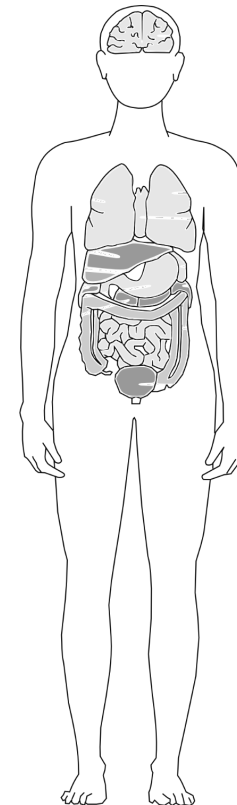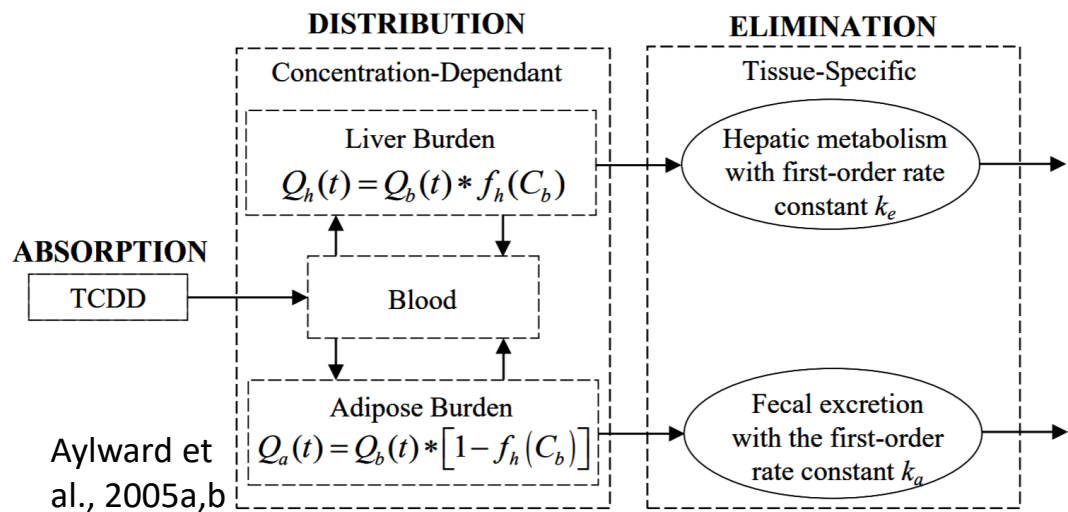
The only PBTK model parameters that need to be estimated for each new chemical are parameters representing chemical-body interactions.

The specifics of these parameters will depend on the details of the PBTK model, but here are a few examples.

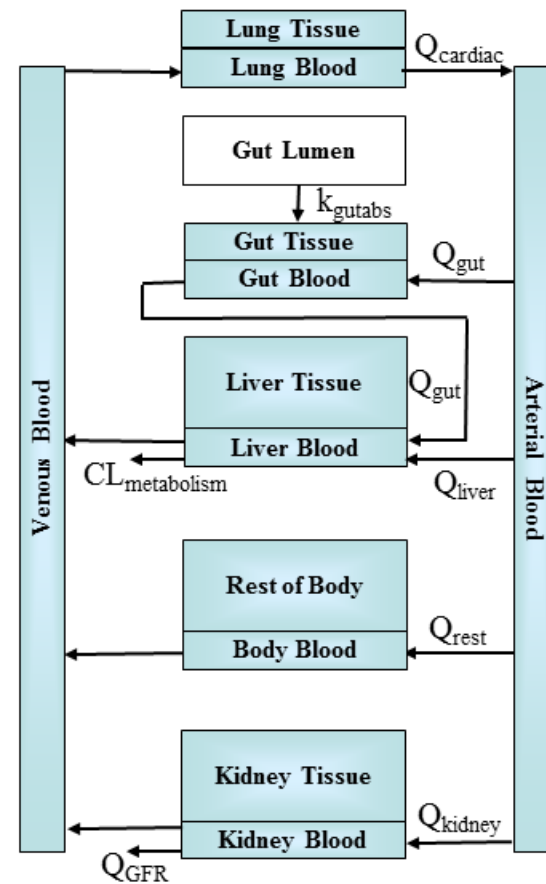| Examples of chemical-specific PBTK model parameters	Explanation
Rate of hepatic metabolism of chemical	How fast does liver break down chemical?
Plasma protein binding	How tightly does the chemical bind to proteins in blood plasma? Liver may not be able to break down chemical that is bound to plasma protein.
Blood:tissue partition coefficients	Assuming chemical diffuses between blood and other tissues very fast compared to the rate of blood flow, the ratio of concentration in blood to concentration in each tissue is approximately constant = partition coefficient.
Rate of active transport into/out of a tissue	If chemical moves between blood and tissues not just by passive diffusion, but by cells actively transporting it in or out of the tissue
Binding to other tissues	Some chemical may be bound inside a tissue and not available for diffusion or transport in/out

PBTK models may be *bespoke* (developed to describe detailed ADME processes for a given chemical) or *generic* (same for all chemicals)

Bespoke model example:
Carrier-Aylward model for TCDD

DISTRIBUTION

Concentration-Dependant

Liver Burden

$$Q_h(t) = Q_b(t) * f_h(C_b)$$

ABSORPTION

TCDD

Blood

Adipose Burden

$$Q_a(t) = Q_b(t) * \left[1 - f_h(C_b)\right]$$

Aylward et al., 2005a,b

ELIMINATION

Tissue-Specific

Hepatic metabolism with first-order rate constant k_e

Fecal excretion with the first-order rate constant k_a

- Describes liver and adipose compartments only – others not relevant for TCDD
- Describes how liver distribution increases as body concentration increases (due to liver binding) – specific to TCDD
- Describes excretion via lipids in feces – again specific to TCDD

Generic model example:
PBTK model in "httk" R package

Lung Tissue
Lung Blood — $Q_{cardiac}$

Gut Lumen
k_{gutabs}

Gut Tissue
Gut Blood — Q_{gut}

Liver Tissue — Q_{gut}
Liver Blood
$CL_{metabolism}$ — Q_{liver}

Rest of Body
Body Blood — Q_{rest}

Kidney Tissue
Kidney Blood — Q_{kidney}
Q_{GFR}

Venous Blood

Arterial Blood

- Compartments are the same for every chemical
- No conc.-dependent tissue binding
- First-order hepatic metabolism for every chemical
- Passive renal clearance for every chemical

Wambaugh et al. (2015)
Pearce et al. (2017a)
Ring et al. (2017)
Linakis et al. (2020)

PBTK models are particularly useful for *extrapolation* (compared to empirical compartmental models)

- Inter-species extrapolation

- Route-to-route extrapolation

- Internal-external extrapolation (reverse TK)

- Extrapolation to different chemicals

- *In vitro-in vivo* extrapolation

Breen et al. (2021)

Inter-species extrapolation: Just substitute in the physiological parameters for another species (e.g., rat liver volume instead of human liver volume)
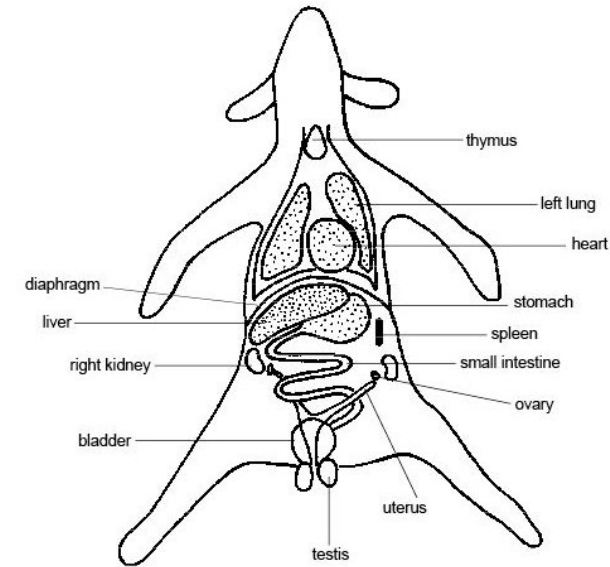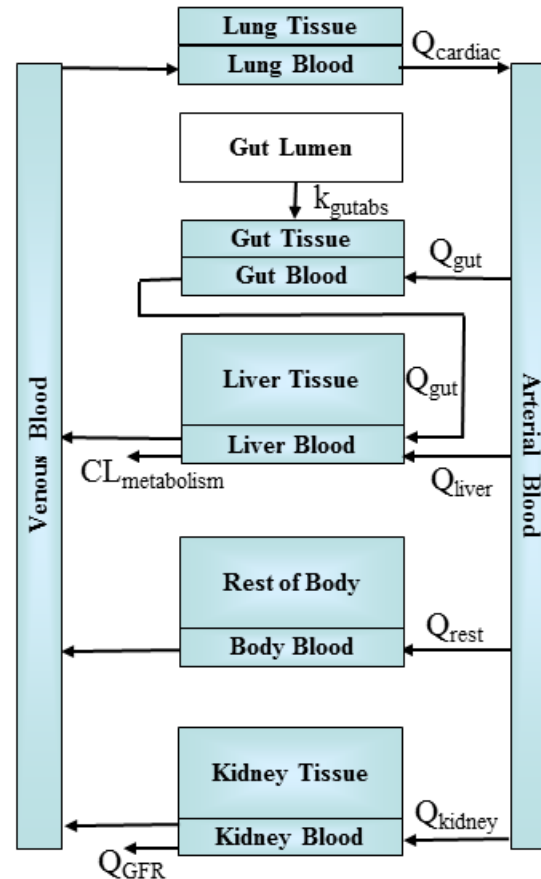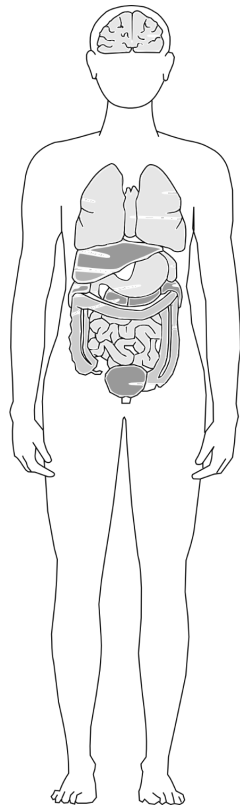
Assumes that chemical-specific parameters stay the same across species – e.g. liver metabolism is the same in rats & humans – but this is often not such a bad assumption

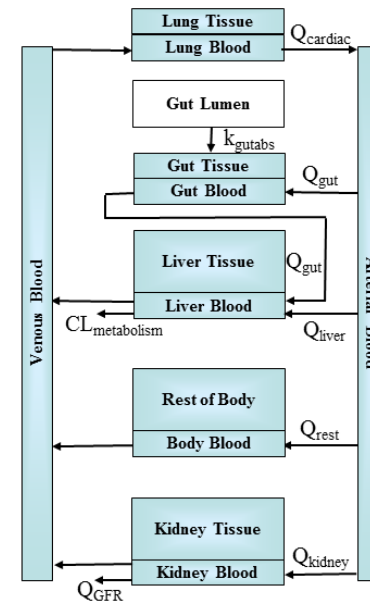# Internal-external extrapolation: Reverse toxicokinetics or reverse dosimetry

Find unknown external exposure?

Known internal dose

| External Exposure | ← | PBTK model | ← | Internal dose |

Tan et al. (2007)

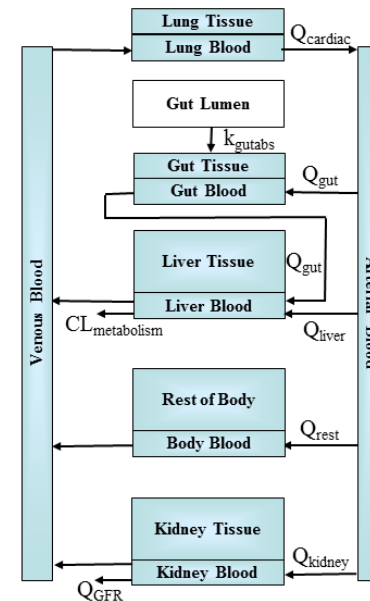# Internal-external extrapolation: Reverse toxicokinetics or reverse dosimetry

Find unknown external exposure?

Known internal dose

| External Exposure | ← | PBTK model | ← | Internal dose |

N.B. PBTK models are complicated, so we typically don't solve *analytically* for external exposure in terms of internal dose.

Instead, we solve *numerically*, e.g. using optimization algorithms.

Lung Tissue
Lung Blood — $Q_{cardiac}$

Gut Lumen
k_{gutabs}
Gut Tissue
Gut Blood — Q_{gut}

Liver Tissue
Q_{gut}
Liver Blood
$CL_{metabolism}$ — Q_{liver}

Rest of Body
Body Blood — Q_{rest}

Kidney Tissue
Kidney Blood — Q_{kidney}
Q_{GFR}

Venous Blood

Arterial Blood

Reverse TK is how the EPA estimated the external exposure of TCDD associated with sperm concentration decrease
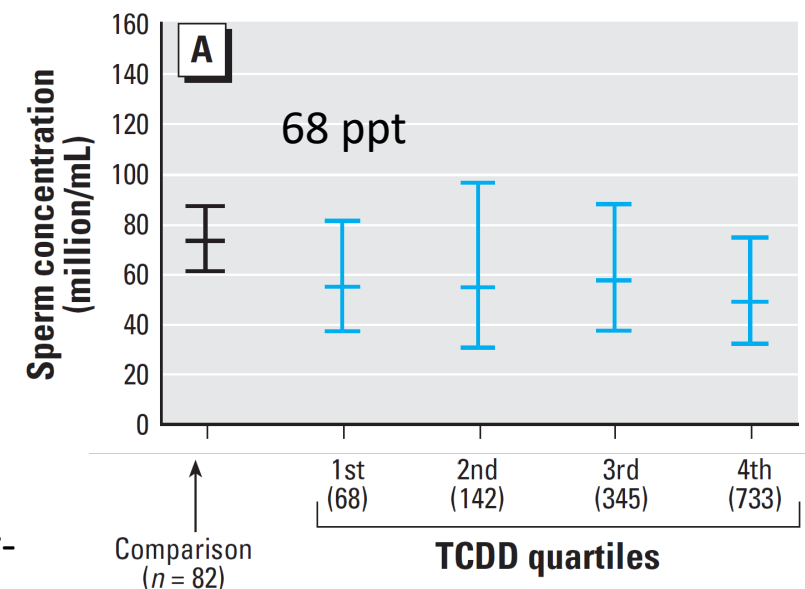
External Exposure ← PBTK model ← Internal dose
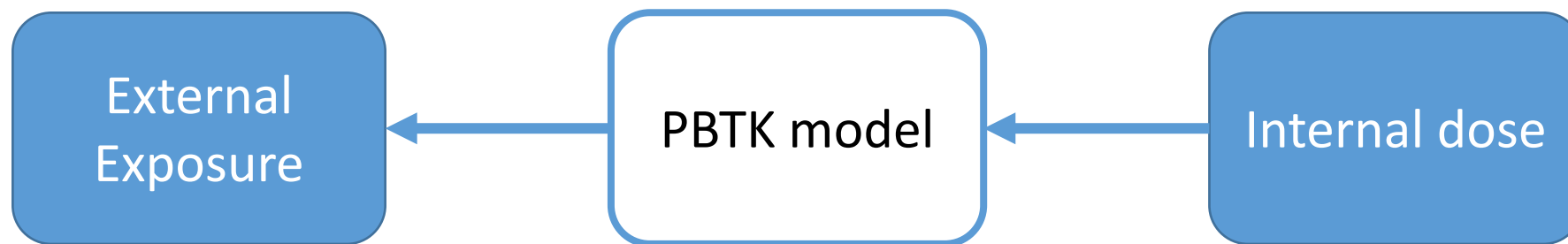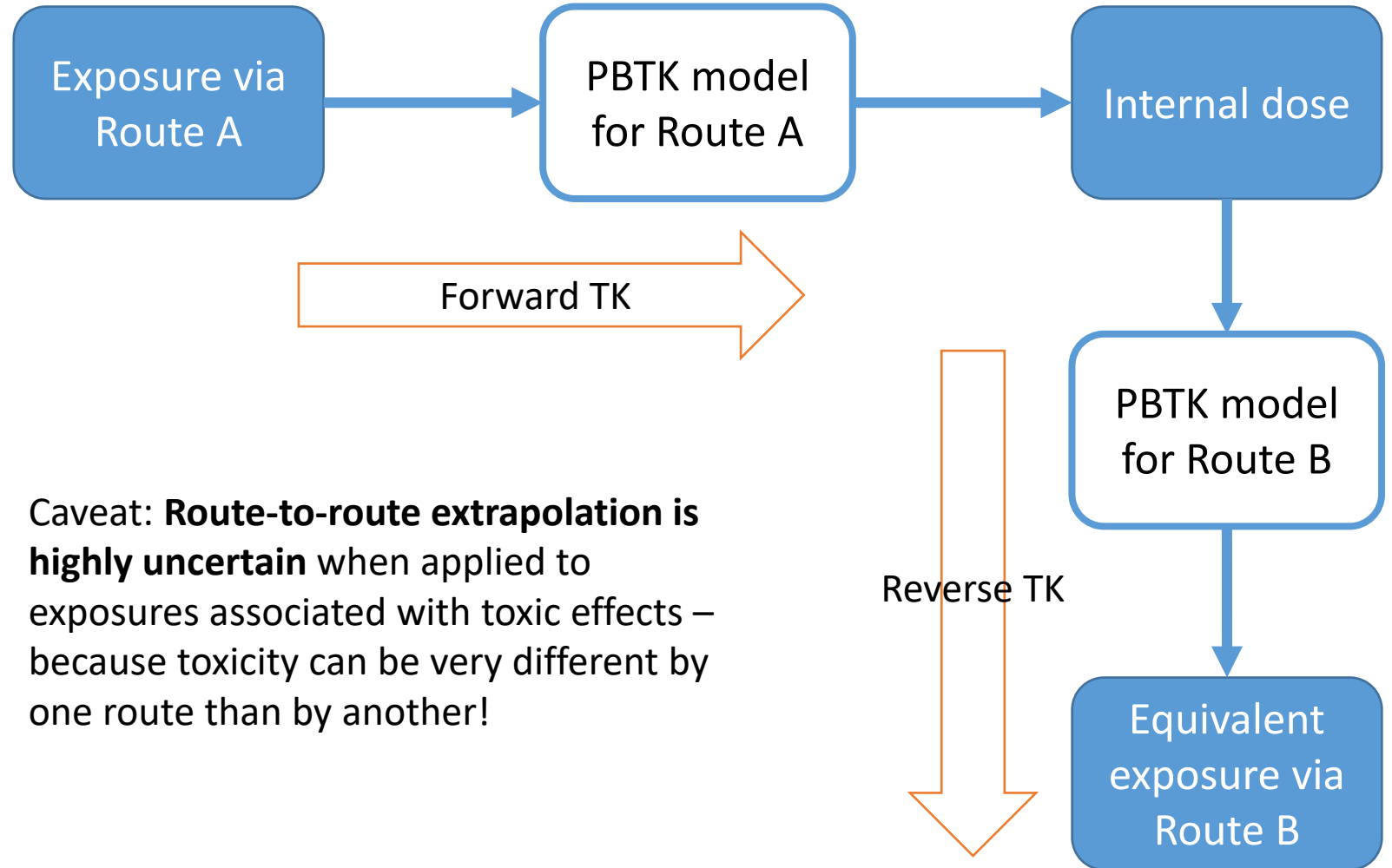
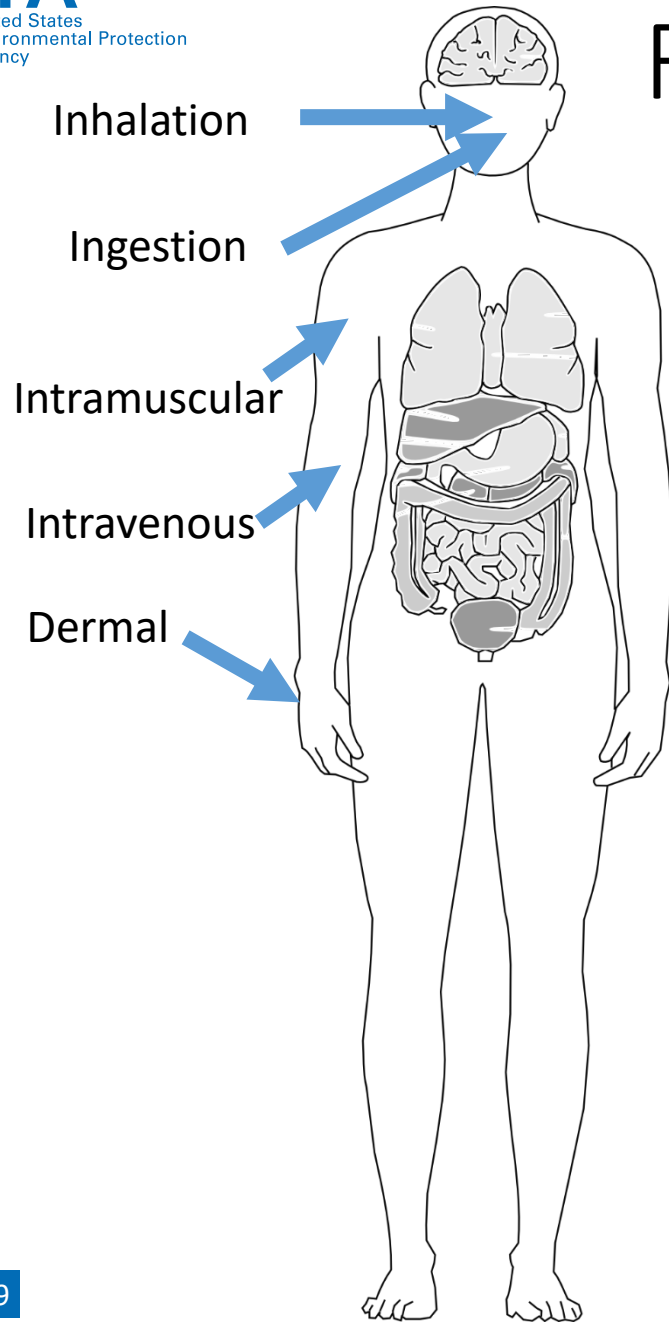0.02 ng/kg/day

US EPA (2012)
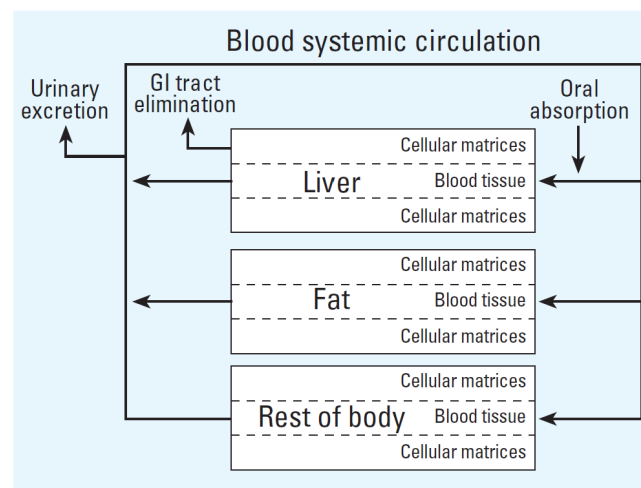
Emond et al. (2005) (similar to Carrier-Aylward, but some differences)


Mocarelli et al. (2008)

Route-to-route extrapolation

Inhalation

Ingestion

Intramuscular

Intravenous

Dermal

Exposure via Route A → PBTK model for Route A → Internal dose

Forward TK

Internal dose → PBTK model for Route B → Equivalent exposure via Route B

Reverse TK

Caveat: **Route-to-route extrapolation is highly uncertain** when applied to exposures associated with toxic effects – because toxicity can be very different by one route than by another!

# Extrapolation to different chemicals: Keep the same generic model structure for all chemicals; just substitute in the different chemical-specific parameters.
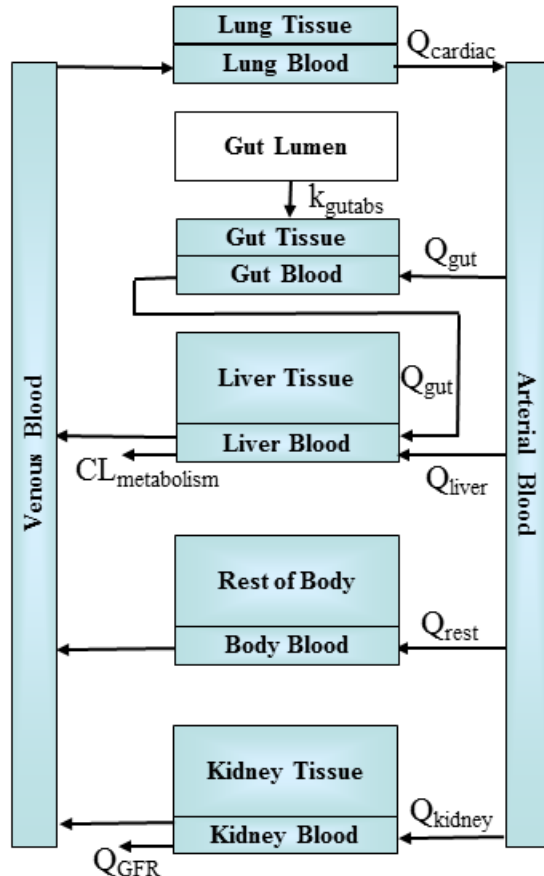
- Same model structure = assume that every chemical undergoes the same *kinds* of processes.
  - E.g., describe hepatic metabolism for all chemicals, but don't describe active transport for any chemicals
- Often it's possible to measure the chemical-specific parameters *in vitro* – can get them rapidly for hundreds of chemicals
- You lose the ability to describe details of specific processes that only pertain to certain chemicals – so your TK predictions might not be exactly correct for each individual chemical.
- But you gain the ability to make reasonably-close TK predictions quickly for large numbers of chemicals! (More on this in the next lecture: *high-throughput toxicokinetics*)

Breen et al. (2012)

In vitro to *in vivo* extrapolation (IVIVE)

Translate *in vitro* bioactive concentration to an equivalent *in vivo* (external) dose. More about this next time!

Adapted from ToxCast Owners Manual

Reverse TK
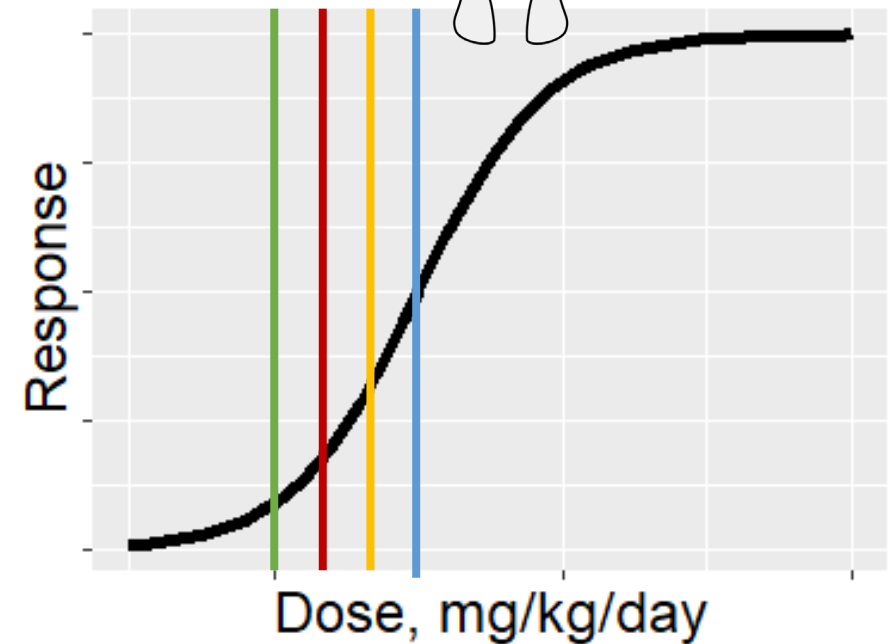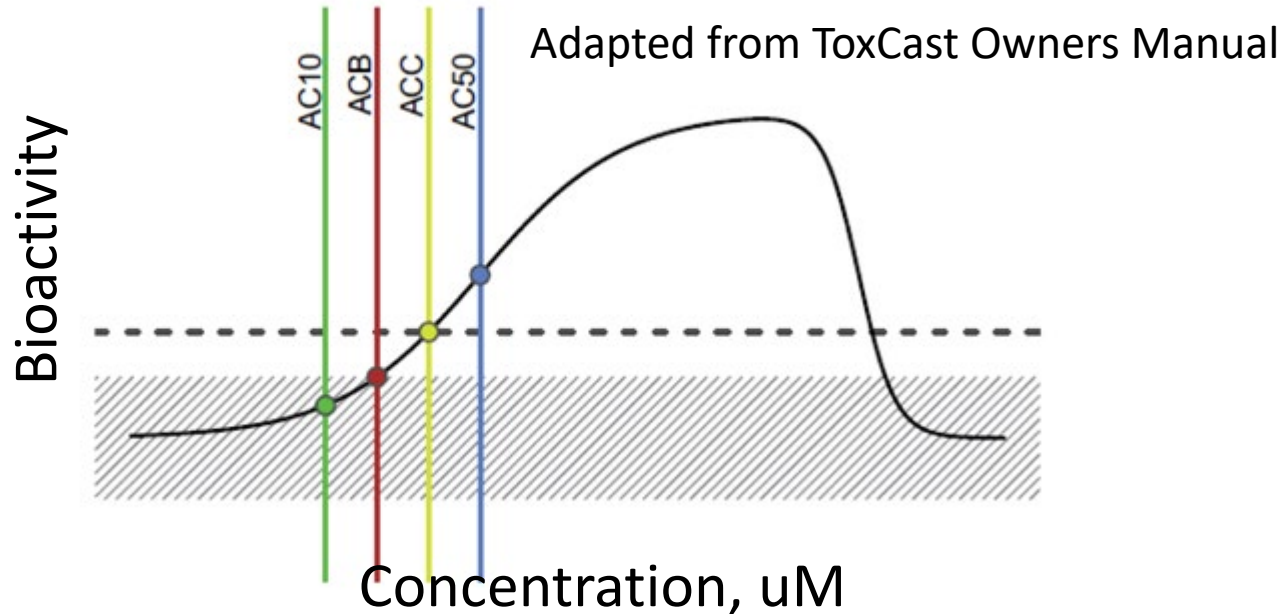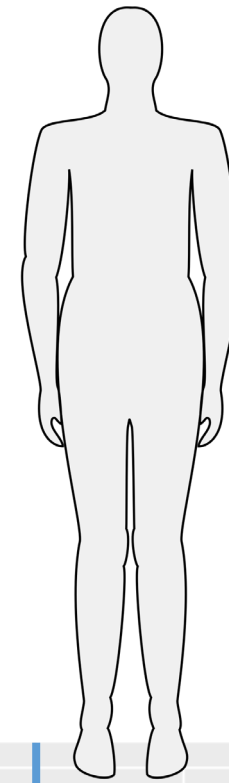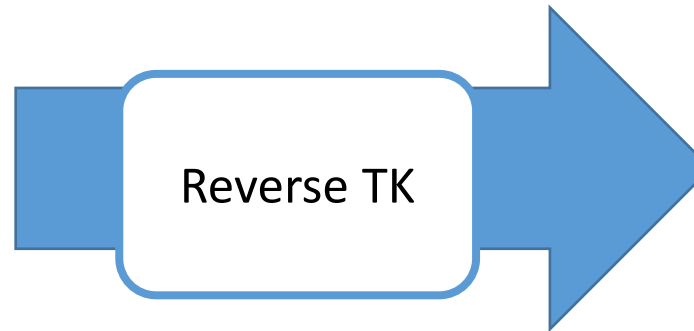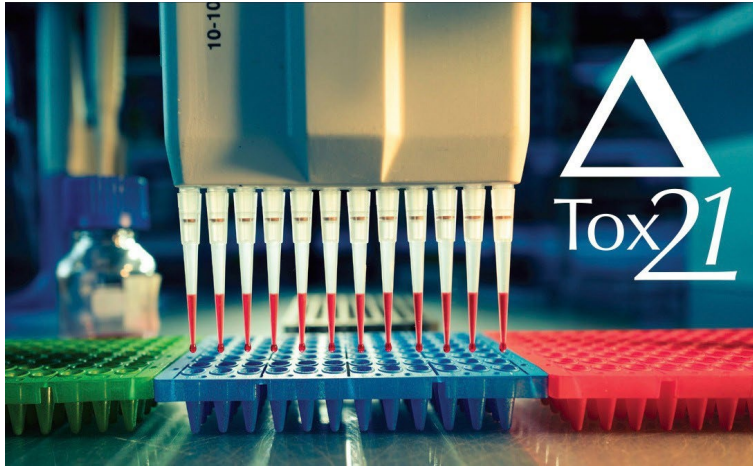
Conclusions

- Toxicokinetics links external exposure to a chemical with internal body concentrations of that chemical by describing ADME
  - Absorption, Distribution, Metabolism, and Excretion of a chemical
- TK modeling can be used to answer questions like "If sperm concentration is reduced when someone has 68 ppt of TCDD in their blood, then what is the corresponding external exposure to TCDD, so EPA can set regulations accordingly?"
- TK models can be simple, empirical models (e.g. 1- or 2-compartment), or more detailed physiologically-based models (PBTK)
- PBTK models may be bespoke (chemical-specific model structure) or generic (same model structure for all chemicals, just different parameters)
- PBTK models are especially useful for extrapolation:
  - Inter-species extrapolation
  - Route-to-route extrapolation
  - Internal-external extrapolation (reverse TK)
  - Extrapolation to different chemicals
  - *In vitro-in vivo* extrapolation

Thank you!

Questions?

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References

1. Aylward, L. L., Brunet, R. C., Carrier, G., Hays, S. M., Cushing, C. A., Needham, L. L., . . . Mocarelli, P. (2005a). Concentration-dependent TCDD elimination kinetics in humans: toxicokinetic modeling for moderately to highly exposed adults from Seveso, Italy, and Vienna, Austria, and impact on dose estimates for the NIOSH cohort. *J Expo Anal Environ Epidemiol, 15*(1), 51-65. doi:10.1038/sj.jea.7500370

2. Aylward, L. L., Brunet, R. C., Starr, T. B., Carrier, G., Delzell, E., Cheng, H., & Beall, C. (2005b). Exposure reconstruction for the TCDD-exposed NIOSH cohort using a concentration- and age-dependent model of elimination. *Risk Anal, 25*(4), 945-956. doi:10.1111/j.1539-6924.2005.00645.x

3. Bourne, D. A. Basic Pharmacokinetics. https://www.boomer.org/c/p4/ Accessed July 31, 2021.

4. Breen, M., Ring, C. L., Kreutz, A., Goldsmith, M. R., & Wambaugh, J. F. (2021). High-throughput PBTK models for in vitro to in vivo extrapolation. *Expert Opin Drug Metab Toxicol, 17*(8), 903-921. doi:10.1080/17425255.2021.1935867

5. Emond, C., Michalek, J. E., Birnbaum, L. S., & DeVito, M. J. (2005). Comparison of the use of a physiologically based pharmacokinetic model and a classical pharmacokinetic model for dioxin exposure assessments. *Environ Health Perspect, 113*(12), 1666-1668. doi:10.1289/ehp.8016

6. Mocarelli, P., Gerthoux, P. M., Patterson, D. G., Jr., Milani, S., Limonta, G., Bertona, M., . . . Needham, L. L. (2008). Dioxin exposure, from infancy through puberty, produces endocrine disruption and affects human semen quality. *Environ Health Perspect, 116*(1), 70-77. doi:10.1289/ehp.10399

7. Tan, Y. M., Liao, K. H., & Clewell, H. J., 3rd. (2007). Reverse dosimetry: interpreting trihalomethanes biomonitoring data using physiologically based pharmacokinetic modeling. J Expo Sci Environ Epidemiol, 17(7), 591-603. doi:10.1038/sj.jes.7500540

8. United States Environmental Protection Agency. (2012). *EPA's Reanalysis of Key Issues Related to Dioxin Toxicity and Response to NAS Comments, Volume 1*. (EPA/600/R-10/038F).