Applying Route-to-Route Extrapolation for Food Ingredients: Considerations & Case Examples

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Conflict of Interest Statement

All views expressed are those of the speaker and do not reflect the views of the FDA or its policies.

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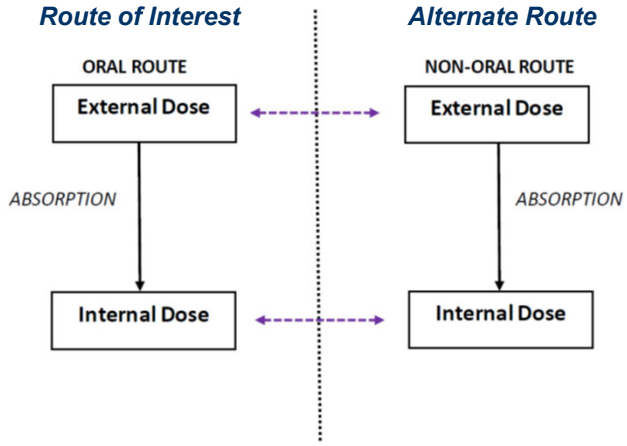
R-to-R Extrapolation



- Extrapolating internal dose from one exposure route to the other
- Predicting effects based on internal dose instead of external exposure levels.

Assumption: Data from studies based on alternate route of exposure (e.g., inhalation, dermal, etc.) are appropriate for use for evaluating safety of a chemical after exposure via the route of interest (e.g., oral).

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R-to-R Extrapolation: Factors

- Physicochemical characteristics: Molecular size, molecular weight, partition coefficient, pKa, solubility, volatility, etc.
- Dosing: Dosing rate, frequency, duration, method of administration, etc.
- Exposure: Contact site, contact duration, contact area, blood flow rate, diffusion barriers

Pharmacokinetics (PK): Absorption, distribution, metabolism (hepatic versus extra-hepatic) and elimination

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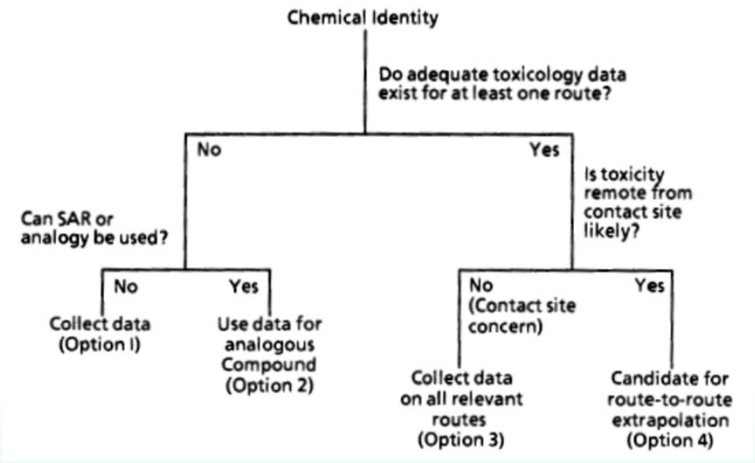
R-to-R Extrapolation: Decision Tree



Gerrity and Henry, 1990

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R-to-R Extrapolation for Food Ingredients

The safety assessment of food ingredients is performed primarily based on oral toxicity data.

Adequate toxicity data from oral exposure studies may not be available or may not be of adequate quality to evaluate the safety of some food ingredients.

R-to-R approach enables utilizing data from non-oral (e.g., inhalation) studies for evaluating effects after oral exposure to a food ingredient.

The first step is to evaluate the relevance of data from non-oral studies for evaluating safety of a chemical after oral exposure–***case-by-case***

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Evaluating Relevance of Non-Oral Data

1. Examining PK equivalence

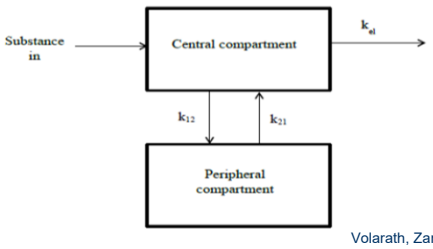
Estimating or comparing internal exposure between the two routes:

- Qualitative – ADME characteristics
- Quantitative – PK parameters (AUC, bioavailability, $t_{1/2}$, t_{ss}, clearance, etc.)

Classical and physiologically based PK (PBPK) models are resourceful tools for examining PK equivalence between exposure routes.

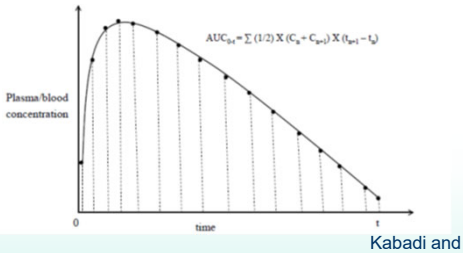
Classical PK Modeling: Brief Overview

- Compartment PK: assumes that the body is a system of one or more compartments



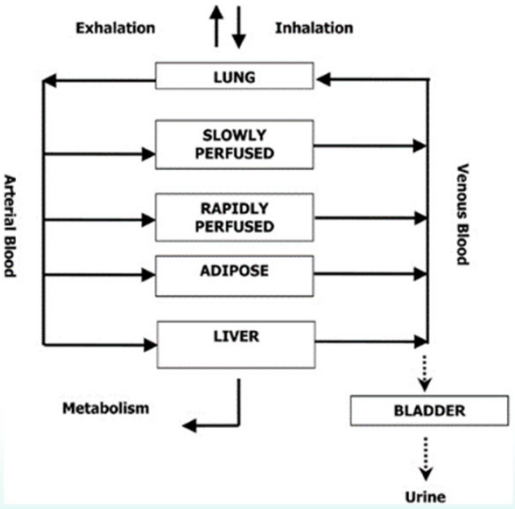
Volarath, Zang and Kabadi, 2019

- Noncompartment PK: assumes that the PK profiles do not depend on number of compartments and are based on estimation of the area under the curve (AUC)



Kabadi and Lin, 2020

PBPK Modeling: Brief Overview



Inserting physiology/anatomy/biochemistry into classical PK:

- Make assumptions
- Build a model
- Run the model
- Validate the model
- Modify/refine the model
- Utilize the model
- Revisit and maintain the model

External dose → Internal dose → Predicting effects

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Evaluating Relevance of Non-Oral Data (continued)

2. Determining toxicological relevance

- Are the effects due to contact (portal of entry) or systemic exposure?
- Are there differences in the type or severity of observed or expected effects?
- Are the effects potentially related to the internal dose?
- Are there any differences in potential mechanisms of action?

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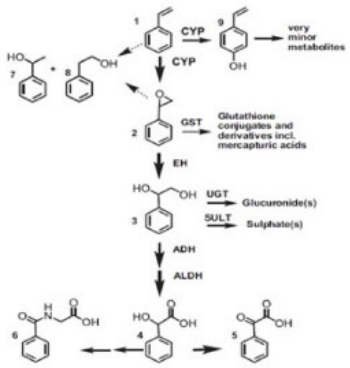
Case Example: Styrene

- Styrene based polymers are regulated for indirect food contact uses.
- Studies after oral and inhalation exposure to styrene have reported increased incidence of tumors in lungs.
  - Is there a need to utilize data from inhalation study to evaluate styrene after oral exposure?
  - If yes, how could the internal dose be calculated?

Human relevance of reported carcinogenic incidences in animal studies of styrene is a subject of debate–beyond the scope of today's discussion

Styrene: Comparing PK between Oral and Inhalation Exposure Routes

- Absorption: 70% (inhalation) and 100% (oral)
- Metabolism: Primarily metabolized by CYPs in the liver and lungs
- Biological half-life: 8-9 hours;
  (first phase with a $t_{1/2}$ of 0.6 hours followed by a second slow phase with a $t_{1/2}$ of 12-13 hours)
- Several PBPK models published on inhalation styrene (including styrene-7,8-oxide (STO)) exposure over the years



Vodicka et al 2008

Styrene: Compare Effects Reported in Available Carcinogenicity Studies

Exposure Route	Oral (NCI, 1979)	Inhalation (Cruzan et al. 2001)
Species/strain	Male and female B6C3F1 mice	Male and female CD-1 mice
Group size	50 mice/group/sex	60 mice/group/sex
Exposure levels	0, 150 and 300 mg/kg bw/d (v/v; in corn oil)	0, 20, 40, 80 and 160 ppm (whole body)
Exposure duration	78 weeks (5d/week)	104 weeks (6h/d; 5d/week)
Carcinogenic incidences (statistically significant)	Bronchioloalveolar adenoma/carcinoma (9/43) vs control (0/20) in males at 300 mg/kg bw/d	Bronchioloalveolar adenoma/carcinoma- 35/50 vs 15/50 in males at 40 ppm, and 16/50 vs 6/50 in females at 20 ppm

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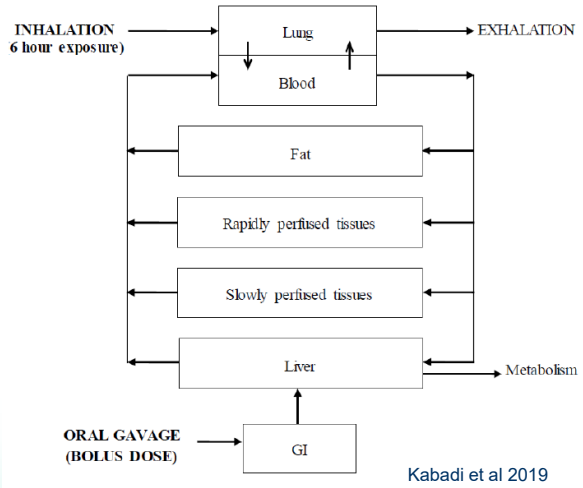
PBPK Modeling of Styrene: Oral and Inhalation



Kabadi et al 2019

- A simple PBPK model of single oral and inhalation styrene exposure was constructed.
- Model was validated using published styrene PK data (Csanady et al 1994 and Filser & Gelbke 2016).
- Changes in levels of styrene in blood versus some tissues were examined.
- All metabolism was evaluated together under liver.
- Metabolites were not examined.

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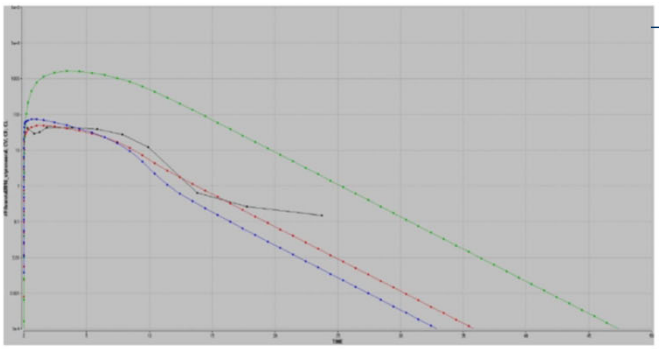
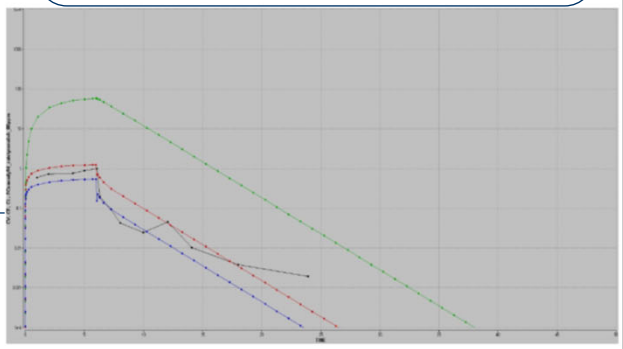
PBPK Model Simulations: Oral versus Inhalation



Oral (300 mg/kg):
Black: experimental (Filser and Gelbke 2016, 500 mg/kg)
Red: Venous Blood
Green: Fat
Blue: Liver



Inhalation (80 ppm):
Black: experimental (Csanady et al 1994)
Red: Venous Blood
Green: Fat
Blue: Liver

Kabadi et al 2019

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Internal Exposure Comparison: Oral versus Inhalation

Exposure route	$AUC_{0\text{-}24\ (observed)}$ ($mgl^{-1}h$)	$AUC_{0\text{-}24\ (predicted)}$ ($mgl^{-1}h$)	$AUC_{(obs/predicted)}$
Inhalation (80 ppm)	4.57	7.68	0.60
Inhalation (600 ppm)	130.08	183.56	0.71
Oral (500 mg/kg)	357.79	809.58	0.44

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PBPK Modeling of Styrene: Implications

- Internal exposure increased with an increase in external exposure; however, metabolism potentially showed saturation at higher exposure levels, irrespective of the exposure route.
- Styrene partitioned into fat more than other tissues across exposure routes.
- The concentrations of styrene in blood and all evaluated tissues declined within 24 hours.

Inhalation data could be used for evaluating styrene after oral exposure.

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Converting Inhalation Exposure into Equivalent Internal Dose

- Inhalation exposure–reported as ppm in air
- Oral exposure–evaluated as equivalent daily dose (mg/kg bw/d)

Converting ppm into equivalent internal dose (based on principles of inhalation dosimetry with inclusion of physiological and PK parameters):

1. Reported inhalation exposure (ppm) → Exposure (mg/ml)
2. Adjusting Exposure for duration → Time-weighted exposure (mg/ml)
3. Time-weighted exposure (mg/ml) → Equivalent daily dose (mg/kg bw/d)
4. Accounting for systemic absorption → Equivalent internal dose (mg/kg bw/d)

This estimated equivalent internal dose could be utilized for calculating a POD.

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Steps for Converting Inhalation Exposure to Equivalent Internal Dose

1. Exposure (mg/ml) = MW X (1/24.45) X [ppm estimate X (1L/10^6L)]

2. Time-weighted Exposure (mg/ml) = Exposure (mg/ml) X (6 hr/24 hr) X (5d/7d)

3. Equivalent Daily Dose (mg/kg bw/d) =
   Time-weighted Exposure (mg/ml) V AVR (ml/min/kg bw) X (60 min/1hr) X (24hr/d)

4. Equivalent Internal Dose (mg/kg bw/d) = F (%) X Equivalent Daily Dose (mg kg bw/d)

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Equivalent Internal Dose Estimates for Inhalation Styrene Exposure

Inhalation Exposure (ppm)	Equivalent Internal Dose (mg/kg bw/d)
20	17.78
40	35.56
80	71.12
160	142.25

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Conversion of Inhalation Exposure to Equivalent Internal Dose: Some Considerations

- This conversion protocol is useful for calculating equivalent internal dose based on inhalation data for volatile solvents (i.e., substances with high vapor pressure).
- It is not applicable to substances that may not fully vaporize upon inhalation exposure.
- It is also not applicable for substances that demonstrate wash in-wash out effect upon inhalation exposure.
- Appropriate physiological values, such as species-specific alveolar ventilation rate parameters, are required for this conversion.

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Case Example: Cobalt Salts (Oral and Inhalation)

Comparing PK profiles of some cobalt salts between oral and inhalation exposure routes

Human PK data on oral versus inhalation exposure of Co (II,III) oxide (*fraction of disposition)

	Inhalation (Foster et al., 1988)		Oral (Christensen et al., 1993)	
Particle Size (Inhalation) /Sex (Oral)	0.8 μm (male only)	1.7 μm (male only)	Male	Female
Mean half-life ($t_{1/2}$, days)	150-250		-	-
Fractional deposition in the lungs (%)	52	78	-	-
Mean urine excretion	0.327*		9.60±6.06 (μmol/L)	30.14±9.51(μmol/L)
Adjusted for creatinine (μmol/μmol)	-	-	1.35±0.94	3.02±2.21
Mean fecal excretion	0.28*		-	-
Blood concentration (μmol/L)	-	-	3.46±2.96	11.18±5.00
Translocation rate (per participant)	0.35±0.07*	0.21±0.07*	-	-
	0.45±0.07*	0.39±0.15*	-	-
Mechanical clearance rate (per participant)	0.04±0.05*	0.07±0.07*	-	-
	0.20±0.10*	0.10±0.09*	-	-

Hung, Smith and Kabadi, 2019 SOT Meeting

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Cobalt Salts: Conclusions

- PK profiles of cobalt and its salts vary with differences in physicochemical properties, such as particle size, ionic charge, solubility, etc.
- Oral bioavailabilities of cobalt chloride and cyanocobalamin are low (approx. 2%). Sufficient inhalation PK data are not available to calculate an estimate for systemic absorption after inhalation exposure, however, systemic absorption after is expected to be low.
- PK profiles of cobalt oxides (II, III) differ between oral and inhalation exposure routes, based on parameters reviewed thus far.
- Additional factors, such as sex may affect PK profiles of cobalt salts, which have not been evaluated yet.

Based on the reviewed information, inhalation data cannot be used for evaluating cobalt salts after oral exposure.

Case Example: Triethanolamine (Oral and Dermal)

Can data from dermal studies be used to evaluate triethanolamine after oral exposure?

- Much higher systemic exposure from dermal application
  - Rapid absorption after oral as well as dermal exposure, but much higher systemic exposure and slower internalization of the dose after dermal exposure
  - 1800-fold increase in dermal dose proportional to approximately 1600-fold increase in AUC
- Differences in severity of expected toxic effects

Based on reviewed data, extrapolating an equivalent internal dose from dermal studies would be an overestimation of its toxic potential after oral exposure.

R-to-R for Food Ingredients (Summary)

Evaluating food ingredients based on non-oral studies involves:

- Examining PK equivalence
- Determining toxicological relevance

For estimating POD using inhalation data, equivalent internal dose (mg/kg bw/d) can be calculated based on principles of inhalation dosimetry and by incorporating species-specific alveolar ventilation rates and systemic absorption.

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R-to-R Extrapolation: Challenges

- Portal of entry effects
- Modes of action
- Effects of factors, such as sex, age which may introduce variability on PK
- Inconsistencies in R-to-R methodologies used by different organizations
- Case-by-case for food ingredients

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