Framework for Establishing an Internal Threshold of Toxicological Concern

Presentation for the In Vitro to In Vivo Extrapolation for High Throughput Prioritization and Decision Making Webinar Series

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Threshold of Toxicological Concern (TTC)

- TTC is a risk assessment tool that establishes acceptable low level exposure values for chemicals with limited toxicological data.
- TTC databases are based on systemic effects after oral exposures.
- Non-cancer TTC databases consist of distributions of chemical specific oral No Observed Adverse Effect Levels (NOAELs).
- Chemicals in existing TTC databases have been categorized using Cramer classification criteria as an indicator of systemic toxicity.
- TTC threshold limits established by identifying a low percentile NOAEL value (e.g. 5th percentile) from the database and applying appropriate uncertainty factors.
Cumulative Distribution of Oral NOAELs
Application of TTC in a Risk Assessment

R. Kroos et al. / Food and Chemical Toxicology 42 (2004) 65–83

1. Is the substance a non-essential metal or metal containing compound, or is it a polyhalogenated-dibenzodioxin, dibenzofuran, or biphenyl?

- NO
- YES

2. Are there structural alerts that raise concern for potential genotoxicity?

- NO
- YES

3. Is the chemical an aflatoxin-like, azoxy, or N-nitroso compound?

- NO
- YES

4. Does estimated intake exceed TTC of 0.15 μg/day?

- NO
- YES

5. Does estimated intake exceed TTC of 1.5μg/day?

- NO
- YES

   Substance would not be expected to be a safety concern

6. Is the compound an organophosphate?

- NO
- YES

7. Does estimated intake exceed TTC of 16μg/day?

- NO
- YES

8. Is the compound in Cramer structural class III?

- NO
- YES

9. Does estimated intake exceed 90μg/day?

- NO
- YES

10. Is the compound in Cramer structural class II?

- NO
- YES

11. Does estimated intake exceed 540μg/day?

- NO
- YES

12. Does estimated intake exceed 1600μg/day?

- NO
- YES

   Substance would not be expected to be a safety concern

Risk assessment requires compound-specific toxicity data

Negligible risk (low probability of a life-time cancer risk greater than 1 in 10^4 – see text)
Internal TTC – Why it’s Relevant

- Multiple situations in risk assessment where it is more appropriate to address internal exposure rather than external dose
  - Metabolism based read-across assessments
    - Tox assessment is based on metabolite(s) for a parent compound that lacks direct tox data (see example later in presentation)
  - Exposure-based waiving of toxicity data
    - Establishing a dermal penetration threshold below which it would not be necessary to have tox data
  - Low level chemical exposure from more than one exposure route
- Partosch (2015) converted external NOAELs to “internal” NOAELs by multiplying by in silico oral bioavailability estimates for each chemical
  - Good initial first steps
  - Still results in an external dose metric
- The need remains for development of an internal TTC utilizing internal exposure metric (e.g. concentration in blood, area under the curve)
Internal TTC Proposed Approach

**CHEMICAL SPECIFIC ADME DATA**

- Existing Literature Data
- Plasma protein binding
- Hepatic clearance
- Renal clearance
- Oral absorption
- Partition coefficients
- In Silico Estimates

**PK MODELING**

**INTERNAL TTC IDENTIFICATION**

- Internal TTC values identified by:
  - Plotting cumulative distribution of Css for Cramer classes
  - Identify 5th percentile Css values
  - Apply appropriate uncertainty factors to 5th percentile Css values

TTC dataset: NOAELs in mg/kg/day

Steady state blood concentration (Css) in animal
Application of Internal TTC to a Risk Assessment

- Risk assessment based on metabolites (e.g. rapid & complete metabolism; SOI produces one metabolite not covered by any analog) OR
- Exposure based waiving of toxicity data

Utilize appropriate modeling method to estimate blood concentration following consumer product exposure scenario

Compare human blood concentration to internal TTC value to determine MoS
Approach to Develop an Internal TTC

- Base modeling on as much compound specific data as possible
- Use *in silico* tools to estimate parameters not found in the literature
- Recommend experimental work only for key chemicals and key parameters
- Focus verification on chemicals that drive the internal TTC threshold
TTC Databases

• Munro et al. (1996)
  – 613 chemicals
  – Species: rat, mouse, rabbit, hamster
  – Routes: gavage, diet, drinking water
  – Durations: subchronic & chronic
  – NOELs identified (mg/kg/day)

• COSMOS project
  – 553 chemicals
  – Species: rat, mouse, dog, primate, rabbit
  – Route: oral
  – Durations: studies ≥ 28 days
  – Chronic NOAELs preferred (mg/kg/day)

http://www.cosmostox.eu/home/welcome/
Literature Search

• Manual PubMed search “pharmacokinetics [chemical name]”
  – Manual review of title and abstracts for papers of interest
  – ~600 papers collected
  – ~60% of TTC chemicals had a paper available
  – Available papers distributed approximately equally across Cramer Classes
  – Manual review of papers needed to extract PK parameters (in process)

• Opportunity for more robust search using analytics approach

• Literature search will help
  – Identify existing ADME data
  – Prioritize what chemicals need more data for modeling
  – Identify in vivo data to support verification of models
In Silico Prediction of Parameters

• Various options for predicting ADME parameters
  – Swiss Institute of Bioinformatics provides summary of software, web services & databases
    http://www.click2drug.org/index.html
  – Multiple published algorithms for different ADME input parameters

• Robust in silico approaches for predicting metabolism are not currently available
  – QSARs developed to date have limited applicability domain
PK Modeling Approaches

• Multiple pharmacokinetic approaches available as options to use in framework

  – $C_{ss}$ equation

    $$C_{ss} = \frac{k_0 \times F}{(GFR \times F_{ub}) + \left[\frac{(Q_l \times F_{ub} \times Cl_{int})}{(Q_l + F_{ub} \times Cl_{int})}\right]}$$

    Wilkinson and Shand (1975)

  – Commercially available generic PBPK models

    • GastroPlus™ (Simulations plus)
    • ADME WorkBench™ (Aegis Technologies)
    • SimCyp™

  – Freely available generic PBPK models
Initial Model Evaluation

• Identify a PK modeling approach with ability to process large batches of chemicals and generate steady state concentrations in blood
  – Batch mode approach needed to support large size of TTC dataset and the number of anticipated loops through the process

• GastroPlus™ and $C_{ss}$ equation

• Chemical specific input parameters (e.g. metabolism, protein binding) were all *in silico* estimates derived from ADMET Predictor™
  – Due to use of all *in silico* input parameters, estimates of $C_{ss}$ are not expected to be quantitatively accurate. Current objective is not to derive accurate estimates of $C_{ss}$, rather to identify approach to be utilized within an internal TTC framework.

• Dosing scenario was representative of the tox study where the NOAEL was derived (e.g. species, dose, route)
Initial Model Evaluation Results

External dose → Kinetic modeling → Internal exposure

Similar results achieved using GastroPlus™
Initial Model Evaluation Results

External dose ➞ Kinetic modeling ➞ Internal exposure

Similar results achieved using GastroPlus™
Fit-for-Purpose Approach

Important to understand amount of conservatism in modeling assumptions/approach

- Oral absorption: (Low)
- Hepatic metabolism: (High)
- Non hepatic systemic clearance: (High)

Overall conservatism:
- Low
- High

Etc.

Graphical representation:
- Representative in vivo tox data
- Non-conservative simulation
- Over conservative simulation
- Balanced simulation

Time

Internal Concentration
# Ex. Metabolism Based Read-across

<table>
<thead>
<tr>
<th>Hypothetical assessment for chemical XYZ</th>
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</thead>
<tbody>
<tr>
<td><strong>Usage scenario</strong></td>
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<tr>
<td><strong>Exposure</strong></td>
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<tr>
<td><strong>Tox data</strong></td>
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<td><strong>Dermal penetration</strong></td>
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<td><strong>Protein binding</strong></td>
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<td><strong>Metabolism</strong></td>
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<tr>
<td><strong>Estimated C\textsubscript{ss}</strong></td>
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<tr>
<td><strong>Risk value for QRA</strong></td>
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</table>
| **QRA** | Internal exposure to XYZ < internal TTC \(\rightarrow\) utilize tox data for metabolite A  
Internal exposure to XYZ > internal TTC \(\rightarrow\) further evaluation needed; possible need for new tox data |

\[ C_{\text{ss}} = \left( \frac{k_0}{(GFR \times F_{ub}) \left[ \frac{(Q_l \times F_{ub} \times CL_{\text{int}})}{(Q_l + F_{ub} \times CL_{\text{int}})} \right]} \right) \times BW \]

\[ k_0 = 0.004 \text{ mg/kg/hr (SCCS (2012) H&Ps), 24 hr exposure, BW = 70 kg, F}_{ub} = 0.8, CL_{\text{int}} = 80 \text{ L/h, Q}_l = 87 \text{ L/h, GFR} = 7.5 \text{ L/h (Davies 1993)} \]
Published Case Study

- Registrants attempted to use metabolism based read-across to support their chemical
  - parent half life in blood ~ 15 minutes
  - PBPK modeling demonstrated that parent AUC was <1% of metabolite AUC following exposure to parent chemical (i.e. predominant systemic exposure is to metabolite)

- Registrants were unable to adequately justify why the low level, short term systemic exposure to the parent would not represent a human safety concern. As such, they had to perform a developmental toxicity study in rodents.

- Availability of an internal TTC may have allowed for comparison of the systemic exposure to an internal exposure threshold.
What Needs to be True for Success

• Clear and transparent documentation
  – Needs to be easily understood by a non-PK expert
  – Well documented so that critical stakeholders can easily understand the strengths and limitations

• Easily reproducible
  – Allows critical stakeholders to have the opportunity to test and become familiar with approach

• Easy access to tools
  – Utilize tools that are easily accessible and available at a reasonable cost to critical stakeholders so that those interested can have a ‘hands-on’ experience

• Publish case studies
  – Case studies that demonstrate the development, progression and utility of the approach may help with its acceptance

• Cross sector collaboration
  – Will increase the diversity in perspectives
Anticipated Challenges

• Predicting if hepatic metabolism is activating or inactivating
  – Will determine if a hepatic metabolism rate is conservative or not
• Other factors that could impact internal concentration
  – Extrahepatic metabolism, renal clearance, transporters
• Regulatory acceptance of PBPK modeling
  – Not all Regulatory agencies have accepted the use of PBPK modeling
• Verification work
  – Limited *in vivo* PK data for comparison to estimates
• Confidence in existing literature data
  – Data from multiple sources over decades will include data that is of poor quality
• Need for additional *in vitro* data
  – Requires time, money, analytical methods
Predicting Impact of Hepatic Metabolism

An example of a preliminary workflow for predicting impact of metabolism

Work by Patra Volarath (former post doc of Ann Richard, US Environmental Protection Agency)

More work is still needed due to limitations of Meteor and Derek
- Many Derek alerts are based on metabolites
- Meteor doesn’t necessarily predict the toxicologically important metabolites
Next Steps for Internal TTC Work

- Proposed internal TTC framework presented at Cosmetics Europe workshop in Sept 2015
- Internal TTC to be a part of the Cosmetics Europe Long Range Science Strategy (LRSS) research program 2016-2020
- Cosmetics Europe working group will form in early 2016 to begin executing internal TTC work
  - Thorough literature search for existing ADME data
  - Identify ADME data gaps for TTC chemicals and selectively generate new in vitro data
  - Evaluate different PK modeling approaches
- There is still a need for strategic partners. If interested contact either:
  - Corie Ellison (ellison.ca@pg.com)
  - Harvey Clewell (hclewell@scitovation.com)
Extending Beyond Internal TTC

The experience gained through this work will be applicable to broader issues as well

- Single chemical PBPK model development
- Balancing conservatism in estimates
- Model verification with limited or no chemical specific in vivo PK data
- Group PBPK read across approaches
- Route to route extrapolation
- In vitro to in vivo extrapolation
ADME WorkBench - http://www.admewb.com/


SimCYP - http://www.simcyp.com/

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