# Skin absorption: defaults, predictions and measurements

*Dr. J.J.M. van de Sandt*

**TNO | Knowledge for business**

# Defaults – predictions - measurements

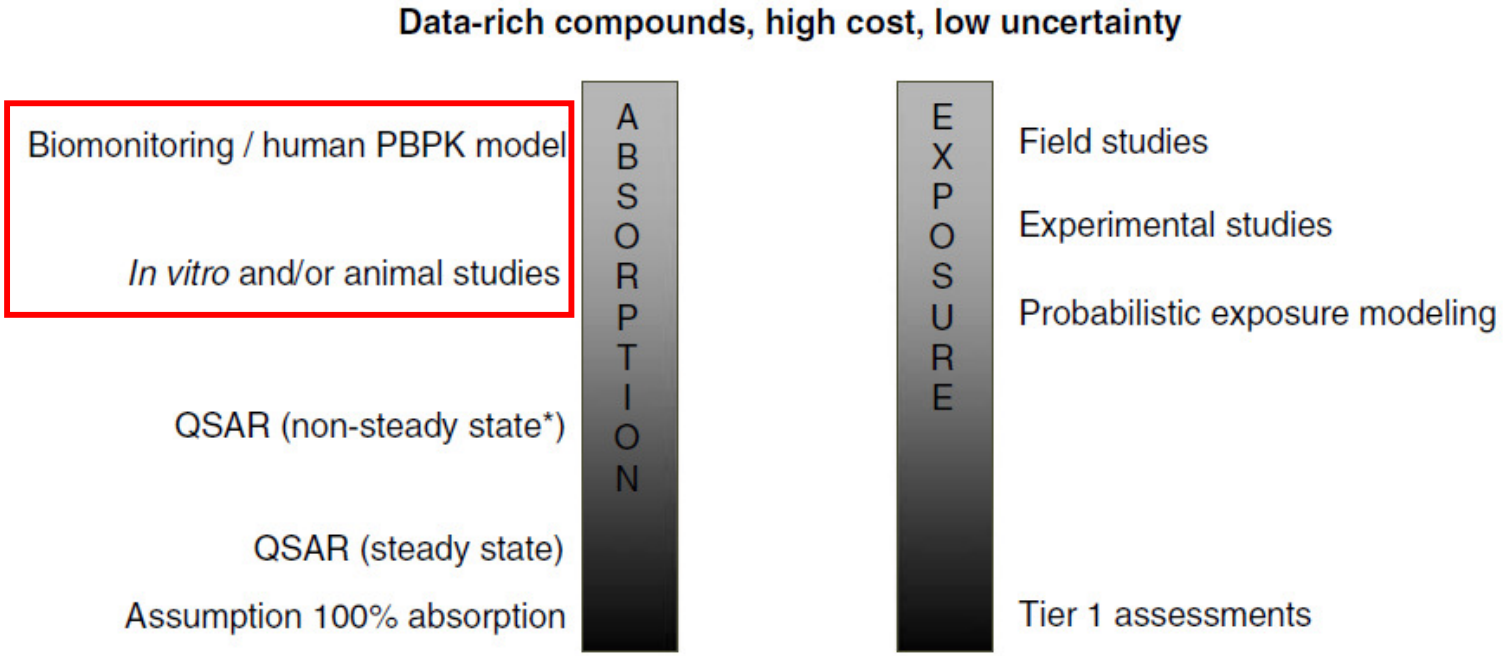
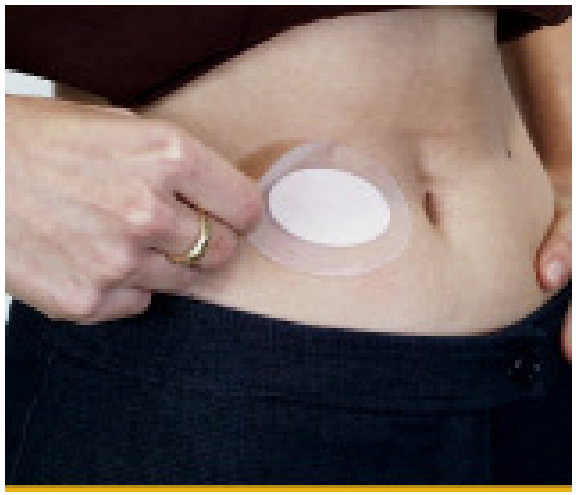


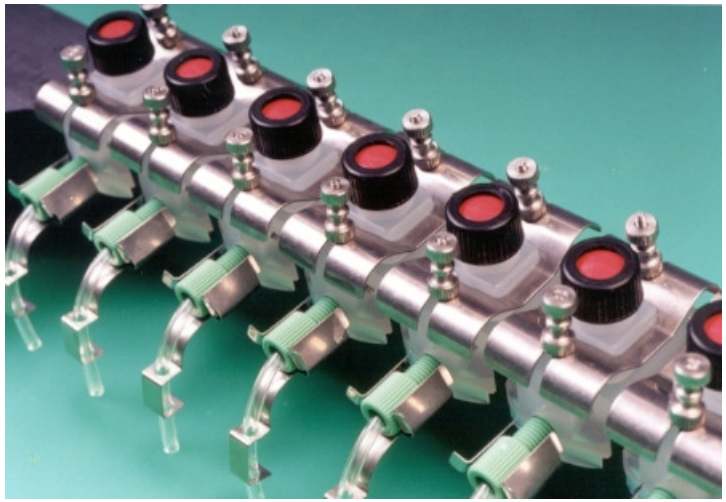

**Figure 1.** Tiered approaches for data generation on skin absorption and exposure. *Non-steady state equations are not available at present.

# Measurements

Animal studies



Volunteer studies



In vitro studies

# Defaults – predictions - measurements

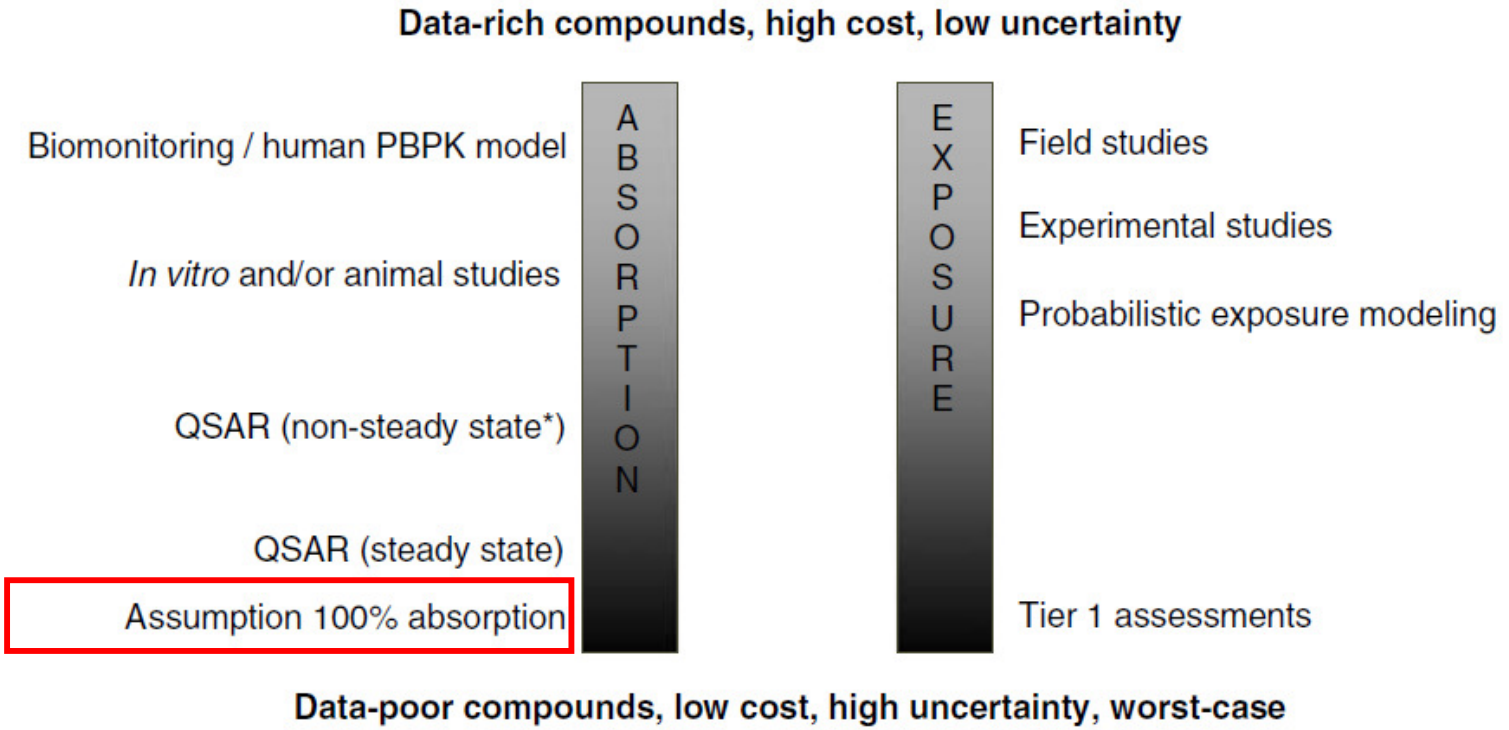



**Figure 1.** Tiered approaches for data generation on skin absorption and exposure. *Non-steady state equations are not available at present.

# Default values for skin absorption

Risk assessment of chemicals within EU (EC, 2004; TGD, 2003):
default of 10% absorption for compounds with:

MW>500, and log Po/w < -1 or > 4

If not: 100% (or testing)

This approach does not take into account:
- Concentration of compound in product
- Vehicle / formulation
- Exposure scenario

# Defaults – predictions - measurements

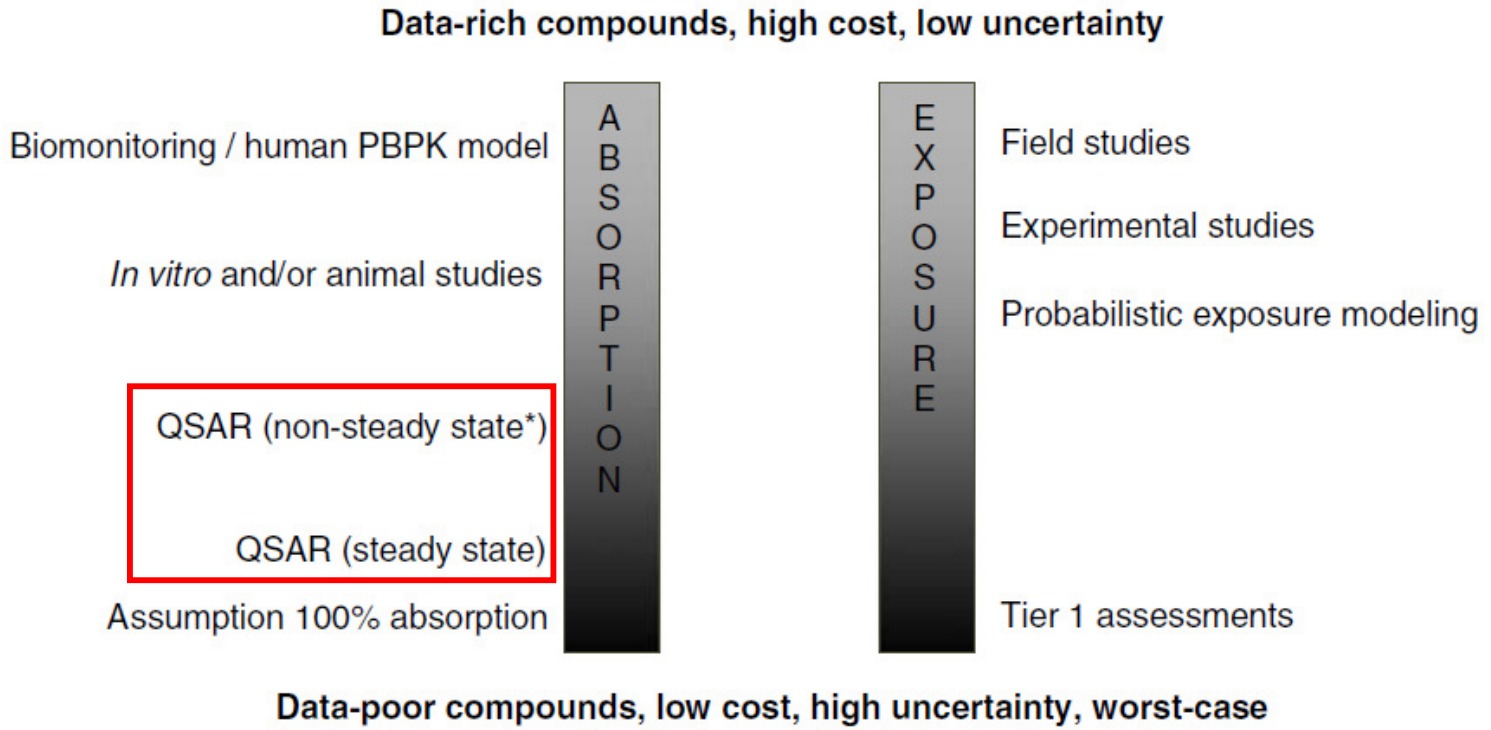



**Figure 1.** Tiered approaches for data generation on skin absorption and exposure. *Non-steady state equations are not available at present.

# Evaluation of QSARs relevant for risk assessment

- Collection of non-proprietary QSARs from public literature and domains
- Evaluation of the selected QSARs based on the OECD principles and practical use
- External evaluation using high quality *in vitro* data

# Evaluation based on OECD criteria

## Criteria for evaluating QSARs for their use in regulatory Risk Assessment

- Properly defined endpoint
- Unambiguous algorithm
- Defined domain of applicability
- Validated (internal and external)
- Mechanistic base

# A funnel approach

1. **Endpoint relevant for Risk Assessment**
   → 21 QSARs
2. **Most evolved QSAR of a similar approach**
   → 14 QSARs
3. **Dataset used to build the QSAR is available**
   → 11 QSARs
4. **Statistical method is described and correct**
   → 11 QSARs
5. **Molecular characteristics in algorithm are available**

**Result → 7 QSARs**

# Selected QSARs

1) $\text{Log } K_p = 0.74 \text{ Log } K_{ow} - 0.0091 \text{ MW} - 2.39$

2) $\text{Log } K_p = 0.0652 \text{ Log } K_{ow} - 0.00603 \text{ MW} - 0.623 \text{ ABSQon} - 0.313 \text{ SsssCH} - 2.30$

3) $\text{Log } K_p = 0.71 \text{ Log } K_{ow} - 0.0061 \text{ MW} - 6.3$

4) $K_p = MW^{-0.6} / \{0.33 + [d_{skin}/(2.4 \times 10^{-6} + 3 \times 10^{-5} K_{ow}^{0.8})]\}$

5) $K_p = 1 / [1/(k_{lip}+k_{pol})+1/k_{aq}]$

6) $\text{Log } J_{max} = - 0.0141 \text{ MW} - 4.52$

7) $\%\text{dermal penetration} = 90.6 - 0.3 \text{ MW}$

# External evaluation of the 7 QSARs

## High quality data from TNO + EDETOX database

## Criteria for data selection:

- Skin absorption measured *in vitro* using human skin
- Compound dissolved in aqueous solution or undissolved (neat)
- Compounds non-ionogenic

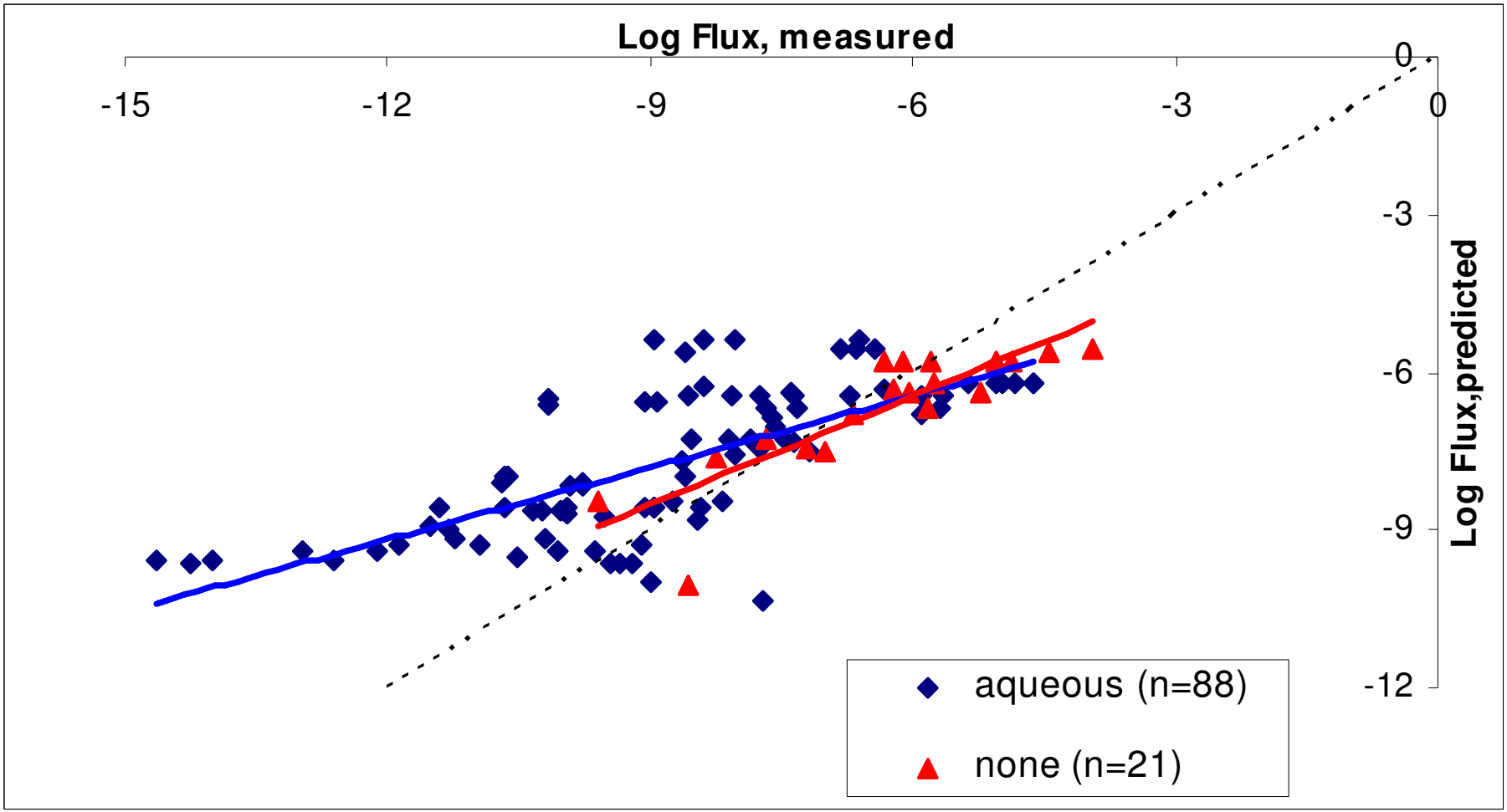
## Result → 70 compounds (100 data points)

# Approach external evaluation

- Predicted values were plotted against measured values to visually and (statistically) investigate the correlation

- Influence of 3 aspects on correlation were examined
  - vehicle (aqueous or none)
  - type of membrane (epidermis or full skin)
  - effect of the (in)finity of the dose applied

# Best performing QSAR (6) (Flux)




All compounds: $y = 0.47\ x - 3.6$; $r^2 = 0.57$

Compounds in water: $y = 0.46\ x - 3.7$; $r^2 = 0.50$

Neat compounds: $y = 0.68\ x - 2.4$; $r^2 = 0.75$

# Overview of most promising (Q)SARs

| *QSAR* | *Applicability domain* | | *External evaluation* |
|---|---|---|---|
| | *MW* | *Log Kow* | |
| 1 | < 350 | between -1 and 4 | $r^2$ < 0.2 |
| 2 | < 350 | between -1 and 4 | $r^2$ < 0.2 (all compounds)<br>**$r^2$ = 0.74 (neat compounds)** |
| 3 | < 500 | between -1 and 5 | $r^2$ < 0.2 |
| 4 | 50 - 200 | Lower than 4 | $r^2$ < 0.2 |
| 5 | < 450 | between 0 and 4 | $r^2$ < 0.2 |
| 6 | < 500 | between -1 and 5 | **$r^2$ = 0.57 (all compounds)**<br>**$r^2$ = 0.75 (neat compounds)** |
| 7 | 150 - 300 | between 4 and 8 | $r^2$ < 0.2 |

# Some conclusions

Parameters predicted by QSARs ($K_p$, $J_{max}$) not used in risk assessment (% of the dose)

Most QSARs did not accurately predict experimentally derived skin penetration data

Potential use of QSARs

- Priority setting
- Infinite dose situations (spills, waste water
- Worst case approach such as TTC concept

# Application of TTC to cosmetic products

TTC values for substances in food should be modified, taking into account:

1. Similarity between cosmetic ingredients and chemical classes from which TTC values for chemicals in food were derived

2. Exposure scenario's

3. Dermal bioavailability
   (route-specific metabolism, skin barrier)

# Bioavailability: route-specific metabolism

Differences in the metabolism of chemicals after oral and topical route of exposure, could result in:

- Increased systemic exposure to the parent compound, following topical exposure, and lower amounts of metabolites

- Decreased systemic exposure to the parent compound, following topical exposure, and higher amounts of metabolites

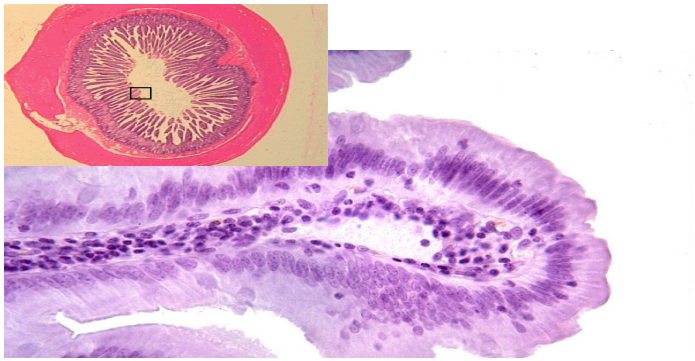
# Bioavailability: route-specific metabolism

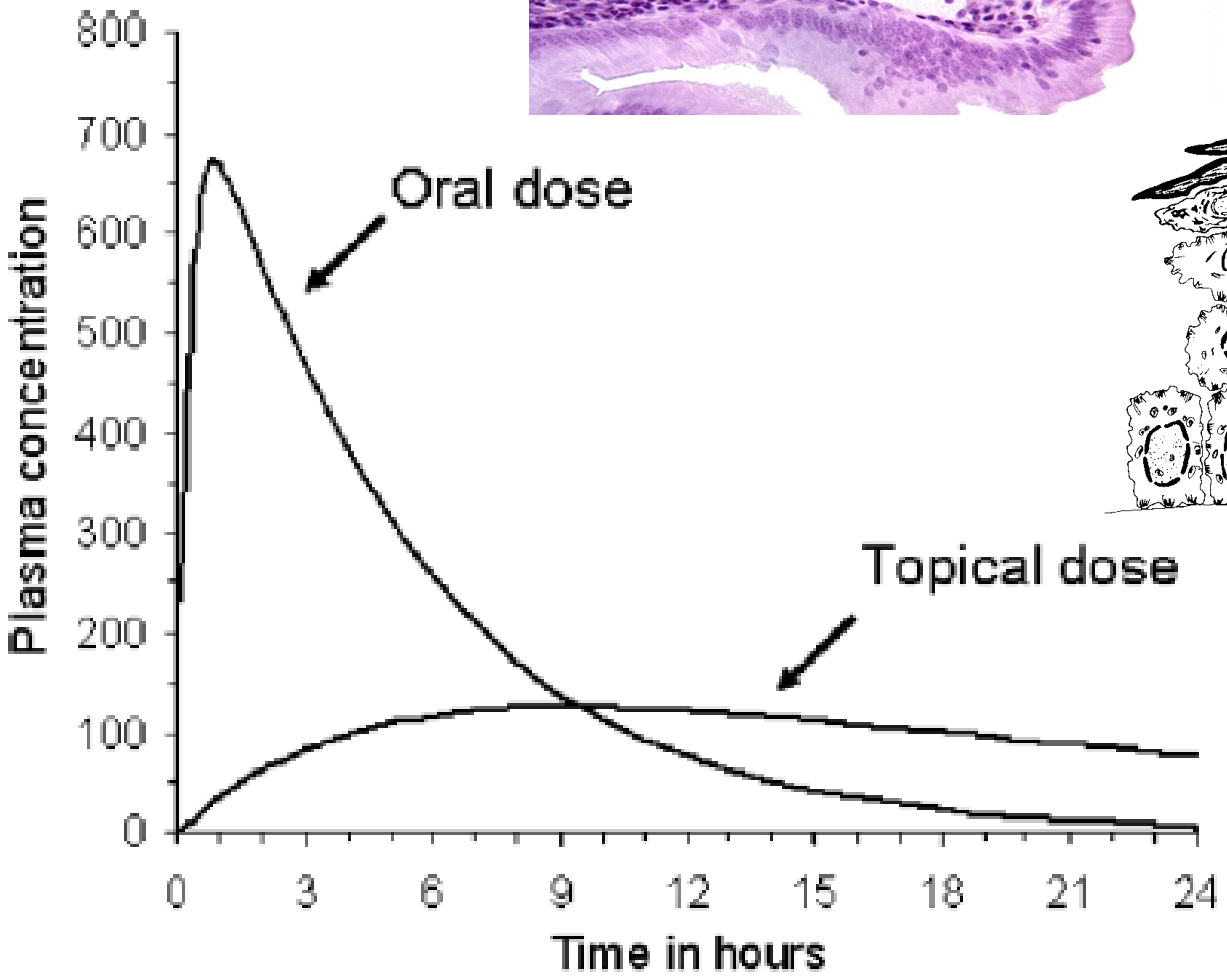
Extensive pre-systemic metabolism is far more likely following oral administration than following topical application

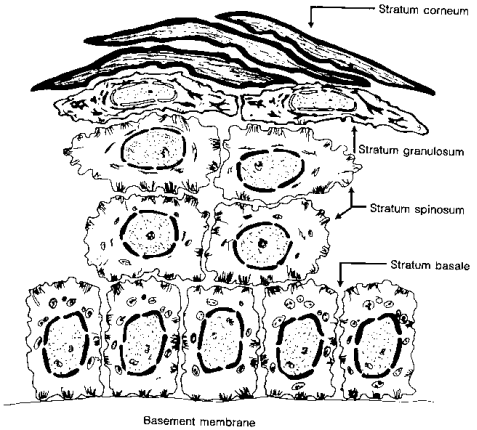
Only under-prediction of potential toxicity following topical application when the toxicity is due to the parent compound and intestinal/hepatic pre-systemic metabolism results in detoxification

Evaluation of pre-systemic of most toxic compounds in all TTC classes indicates that these compounds do not undergo detoxification following oral dosing

# Bioavailability





# EDETOX database

In vitro absorption across human skin (n = 62)

**Log P**: experimental, or ClogP

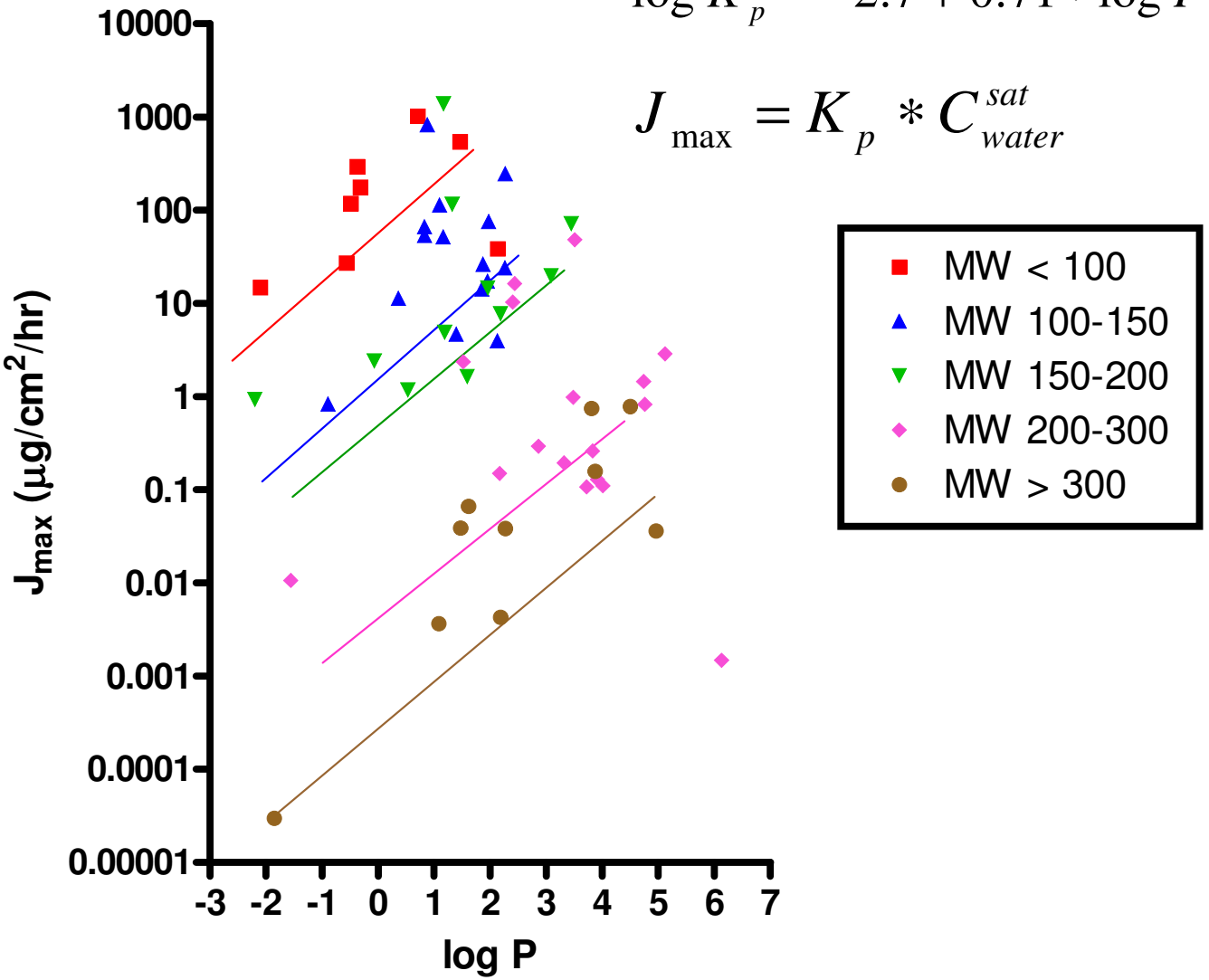
**$C_{sat}$(aq)**: experimental or calculated

| Chemical Name | CAS | MW | LogP | log Kp | Kp (cm/h) | Csat (mg/cm3) | Jmax (mg/cm2/h) | Jmax (µg/cm2/h) |
|---|---|---|---|---|---|---|---|---|
| Methotrexate | 59-05-2 | 454.45 | -1.85 | -6.786 | 1.63E-07 | 0.17 | 2.78E-08 | 0.00003 |
| Benzo[a]pyrene | 50-32-8 | 252.32 | 6.13 | 0.113 | 1.30E+00 | 1.14E-06 | 1.48E-06 | 0.00148 |
| Aldosterone | 52-39-1 | 360.44 | 1.08 | -4.132 | 7.37E-05 | 0.05 | 3.68E-06 | 0.00368 |
| Griseofulvin | 126-07-8 | 352.77 | 2.18 | -3.304 | 4.96E-04 | 8.66E-03 | 4.29E-06 | 0.00429 |
| Acyclovir | 59277-89-3 | 225.21 | -1.56 | -5.181 | 6.57E-06 | 1.62 | 1.06E-05 | 0.01065 |
| Chlorpyrifos | 2921-88-2 | 350.59 | 4.96 | -1.317 | 4.82E-02 | 7.50E-04 | 3.61E-05 | 0.03613 |
| T2 Toxin | 21259-20-1 | 466.57 | 2.27 | -3.934 | 1.16E-04 | 0.33 | 3.83E-05 | 0.03832 |
| Cortisone | 53-06-5 | 360.46 | 1.47 | -3.855 | 1.39E-04 | 0.28 | 3.90E-05 | 0.03903 |
| Hydrocortisone | 50-23-7 | 362.47 | 1.61 | -3.768 | 1.70E-04 | 0.39 | 6.64E-05 | 0.06644 |
| Lindane | 58-89-9 | 290.83 | 3.72 | -1.833 | 1.47E-02 | 7.31E-03 | 1.07E-04 | 0.10733 |
| Estradiol | 50-28-2 | 272.37 | 4.01 | -1.514 | 3.06E-02 | 3.60E-03 | 1.10E-04 | 0.11007 |
| Methylene-Bis-(2-Chloroaniline) | 101-14-4 | 267.00 | 3.91 | -1.553 | 2.80E-02 | 4.62E-03 | 1.29E-04 | 0.12935 |
| Dinitrochlorobenzene | 97-00-7 | 202.55 | 2.17 | -2.395 | 4.02E-03 | 0.037 | 1.49E-04 | 0.14891 |
| Progesterone | 57-83-0 | 314.45 | 3.87 | -1.870 | 1.35E-02 | 1.17E-02 | 1.58E-04 | 0.15754 |
| Testosterone | 58-22-0 | 288.40 | 3.32 | -2.102 | 7.90E-03 | 2.46E-02 | 1.94E-04 | 0.19432 |
| Parathion | 56-38-2 | 291.26 | 3.83 | -1.757 | 1.75E-02 | 1.49E-02 | 2.60E-04 | 0.26031 |
| Parathion methyl | 298-00-0 | 263.21 | 2.86 | -2.275 | 5.30E-03 | 5.50E-02 | 2.92E-04 | 0.29172 |
| Diazinon | 333-41-5 | 304.35 | 3.81 | -1.851 | 1.41E-02 | 5.29E-02 | 7.44E-04 | 0.74420 |
| Butachlor | 23184-66-9 | 311.86 | 4.50 | -1.407 | 3.91E-02 | 2.01E-02 | 7.86E-04 | 0.78631 |
| Triclosan | 3380-34-5 | 289.55 | 4.76 | -1.087 | 8.19E-02 | 1.00E-02 | 8.19E-04 | 0.81875 |
| Fluorouracil | 51-21-8 | 130.08 | -0.89 | -4.125 | 7.48E-05 | 11.07 | 8.28E-04 | 0.82797 |
| Mannitol | 69-65-8 | 182.17 | -2.20 | -5.373 | 4.22E-06 | 220 | 9.29E-04 | 0.92943 |
| Propranolol | 525-66-6 | 257.34 | 3.48 | -1.799 | 1.59E-02 | 6.17E-02 | 9.79E-04 | 0.97946 |
| Nitro-1,4-Benzenediamine | 5307-14-2 | 153.14 | 0.53 | -3.258 | 5.52E-04 | 2.14 | 1.18E-03 | 1.18025 |
| Cinnamyl anthranilate | 87-29-6 | 253.30 | 4.74 | -0.880 | 1.32E-01 | 1.10E-02 | 1.45E-03 | 1.45045 |
| Methylenedianiline | 101-77-9 | 198.27 | 1.59 | -2.781 | 1.66E-03 | 0.99 | 1.64E-03 | 1.63903 |
| Propoxur | 114-26-1 | 209.25 | 1.52 | -2.897 | 1.27E-03 | 1.86 | 2.35E-03 | 2.35378 |
| Caffeine | 58-08-2 | 194.20 | -0.07 | -3.934 | 1.16E-04 | 20.8 | 2.42E-03 | 2.41565 |
| Pentachlorophenol | 87-86-5 | 266.34 | 5.12 | -0.689 | 2.04E-01 | 1.40E-02 | 2.86E-03 | 2.86108 |
| Cinnamic acid | 621-82-9 | 148.16 | 2.13 | -2.091 | 8.09E-03 | 0.49 | 3.97E-03 | 3.96591 |
| Coumarin | 91-64-5 | 146.15 | 1.39 | -2.605 | 2.48E-03 | 1.88 | 4.67E-03 | 4.66738 |
| Acetylsalicylic Acid | 50-78-2 | 180.16 | 1.19 | -2.954 | 1.11E-03 | 4.42 | 4.91E-03 | 4.90698 |
| DEET | 134-62-3 | 191.28 | 2.18 | -2.319 | 4.79E-03 | 1.62 | 7.76E-03 | 7.76406 |
| Nicotinate benzyl | 94-44-0 | 213.24 | 2.40 | -2.297 | 5.04E-03 | 2.04 | 1.03E-02 | 10.29088 |
| Nicotinic Acid | 59-67-6 | 123.11 | 0.36 | -3.195 | 6.37E-04 | 17.8 | 1.13E-02 | 11.33635 |
| Nitrobenzene | 98-95-3 | 123.11 | 1.85 | -2.137 | 7.28E-03 | 1.95 | 1.42E-02 | 14.19641 |
| Methyl-4-hydroxybenzoate | 99-76-3 | 152.14 | 1.96 | -2.236 | 5.80E-03 | 2.53 | 1.47E-02 | 14.66437 |
| Urea | 57-13-6 | 60.10 | -2.11 | -4.565 | 2.72E-05 | 550 | 1.50E-02 | 14.95651 |
| Lidocaine | 137-58-6 | 234.34 | 2.44 | -2.397 | 4.00E-03 | 4.07 | 1.63E-02 | 16.29628 |
| Cinnamyl alcohol | 104-54-1 | 134.18 | 1.95 | -2.134 | 7.34E-03 | 2.36 | 1.73E-02 | 17.31927 |
| Phenylphenol | 90-43-7 | 170.21 | 3.09 | -1.544 | 2.85E-02 | 0.69 | 1.97E-02 | 19.68747 |
| Salicylic acid | 69-72-7 | 138.12 | 2.26 | -1.938 | 1.15E-02 | 2.09 | 2.41E-02 | 24.09157 |
| Benzoic Acid | 65-85-0 | 122.10 | 1.87 | -2.117 | 7.63E-03 | 3.44 | 2.62E-02 | 26.24623 |
| Dimethylnitrosamine | 62-75-9 | 74.08 | -0.57 | -3.557 | 2.77E-04 | 98.5 | 2.73E-02 | 27.30281 |
| Benzene | 71-43-2 | 78.12 | 2.13 | -1.664 | 2.17E-02 | 1.79 | 3.88E-02 | 38.75713 |
| Nicotinate hexyl | 23597-82-2 | 207.27 | 3.51 | -1.472 | 3.37E-02 | 1.43 | 4.82E-02 | 48.17522 |
| Phenoxyethanol | 122-99-6 | 138.17 | 1.16 | -2.719 | 1.91E-03 | 26.9 | 5.13E-02 | 51.28913 |
| Nicotinate methyl | 93-60-7 | 137.14 | 0.83 | -2.947 | 1.13E-03 | 47.6 | 5.37E-02 | 53.68116 |
| Butoxyethanol | 111-76-2 | 118.18 | 0.83 | -2.832 | 1.47E-03 | 44.9 | 6.61E-02 | 66.09036 |
| Safrole | 94-59-7 | 162.19 | 3.45 | -1.240 | 5.75E-02 | 1.24 | 7.13E-02 | 71.34101 |
| Chloroform | 67-66-3 | 119.38 | 1.97 | -2.030 | 9.34E-03 | 8.07 | 7.53E-02 | 75.33379 |
| Benzyl Alcohol | 100-51-6 | 108.13 | 1.10 | -2.579 | 2.64E-03 | 43.1 | 1.14E-01 | 113.61080 |
| Nicotinate ethyl | 614-18-6 | 151.17 | 1.32 | -2.685 | 2.06E-03 | 56 | 1.16E-01 | 115.54927 |
| Methoxypropan-2-ol | 107-98-2 | 90.12 | -0.49 | -3.598 | 2.52E-04 | 470 | 1.19E-01 | 118.52209 |
| Ethoxyethanol | 110-80-5 | 90.12 | -0.32 | -3.477 | 3.33E-04 | 530 | 1.76E-01 | 176.48888 |
| Nicotinate butyl | 6938 06-3 | 129.22 | 2.27 | -1.877 | 1.33E-02 | 18.35 | 2.44E-01 | 243.64412 |
| Dimethylamine | 124-40-3 | 45.10 | -0.38 | -3.245 | 5.68E-04 | 520 | 2.95E-01 | 295.46677 |
| Phenol | 108-95-2 | 94.11 | 1.46 | -2.237 | 5.78E-03 | 94.1 | 5.44E-01 | 544.14601 |
| Catechol | 120-80-9 | 110.11 | 0.88 | -2.747 | 1.79E-03 | 460 | 8.23E-01 | 822.98490 |
| Dimethylethylamine | 598-56-1 | 73.14 | 0.70 | -2.649 | 2.24E-03 | 460 | 1.03E+00 | 1030.70071 |
| Nicotine | 54-11-5 | 162.23 | 1.17 | -2.859 | 1.38E-03 | 1000 | 1.38E+00 | 1382.23488 |

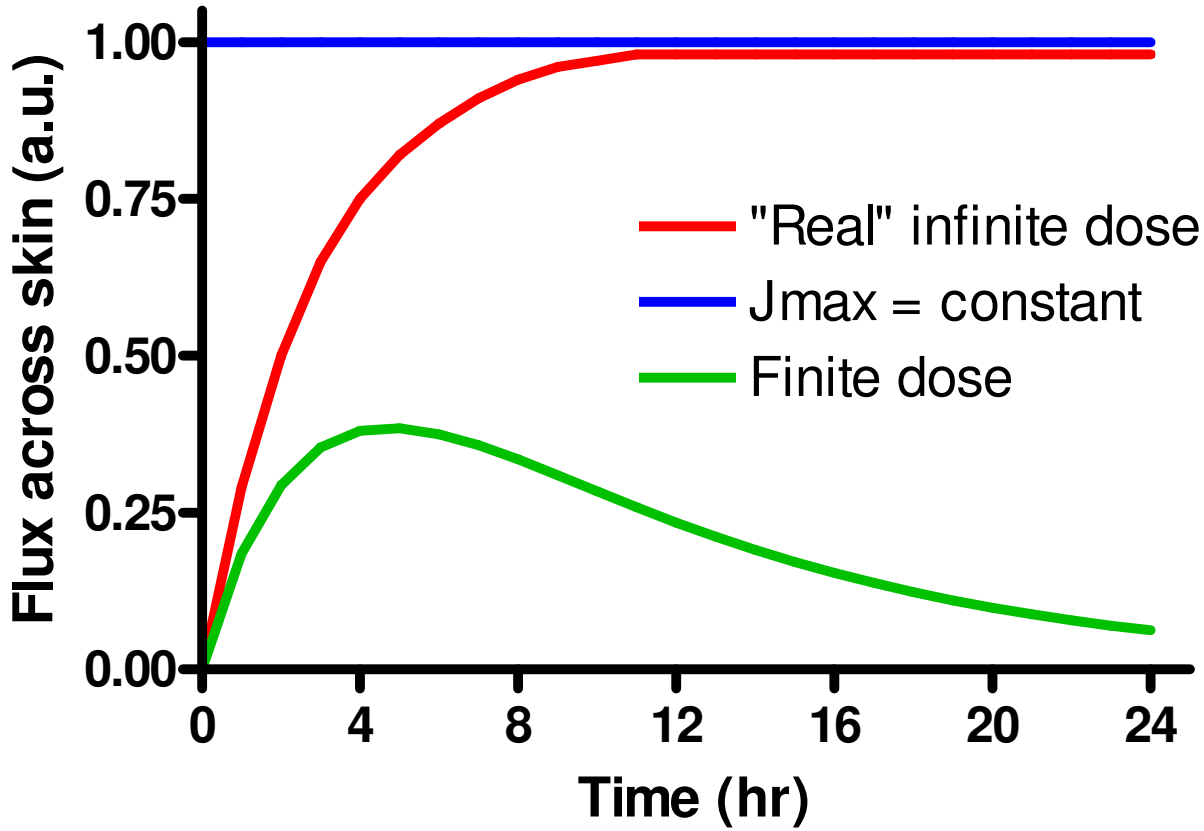
# Edetox maximum fluxes

$$\log K_p = -2.7 + 0.71 * \log P - 0.0061 * MW$$

$$J_{\max} = K_p * C_{water}^{sat}$$



| $J_{max}$ (µg/cm$^2$/hr) | MW | log P |
|---|---|---|
| $J_{max} < 0.1$ | MW > 300 | log P < -1; log P > 5 |
| $0.1 < J_{max} < 1$ | MW = 200-300 | log P > 2, 2.5 |
| $1 < J_{max} < 10$ | MW = 150-250 | log P = 1.0 – 2.0 |
| $10 < J_{max} < 100$ | MW = 60-200 | log P = 0.5 - 3.0 (3.5) |
| $J_{max} > 100$ | MW < 150 | log P = -0.5 – 1.5 (2.0) |


Flux across skin (a.u.)
1.00
0.75
0.50
0.25
0.00
0
4
8
12
16
20
24
Time (hr)
"Real" infinite dose
Jmax = constant
Finite dose

# Absorption under in-use conditions

- Open literature
  - limited number of publications

- SCCNFP opinions
  - Variable quality of data
  - Limited product variability (hair dyes)

- EDETOX database
  - Limited data on cosmetics

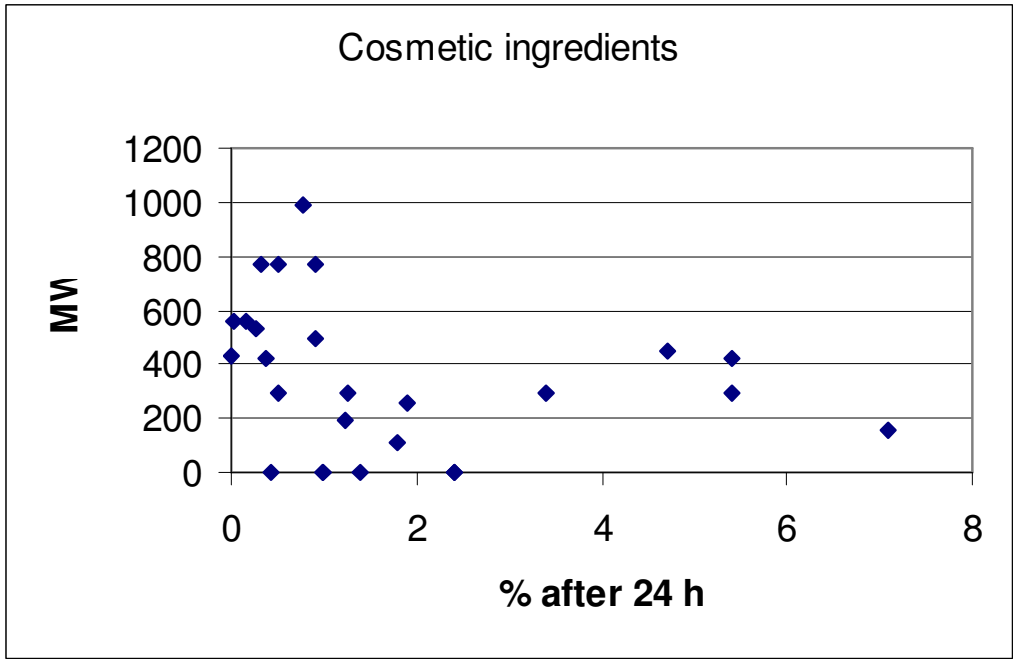
- TNO database
  - Coded compounds

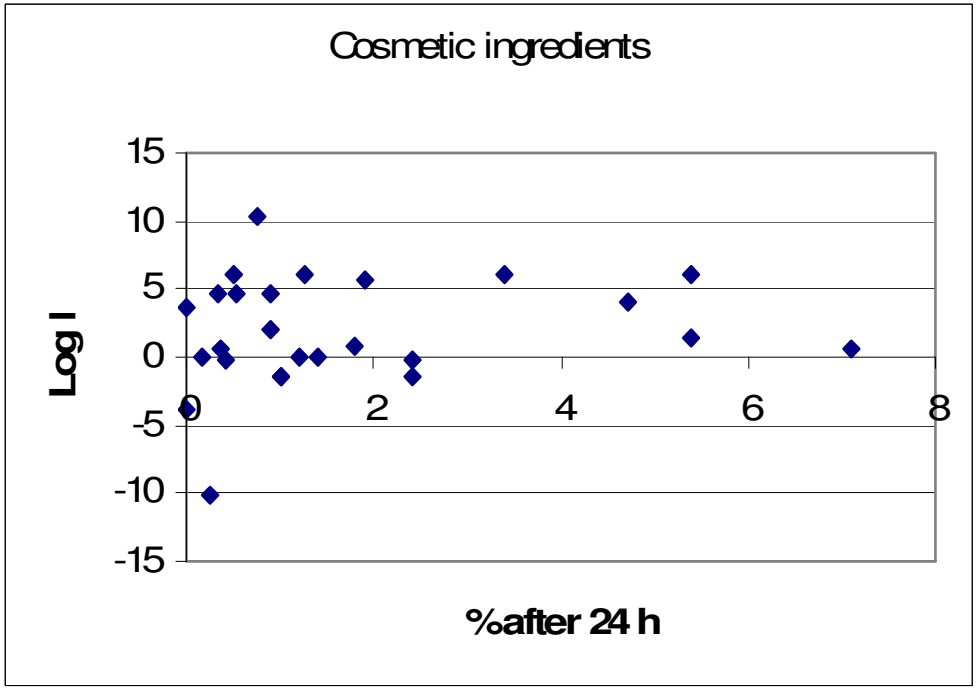
# Absorption under in-use conditions

| | |
|---|---|
| - Exposure period | Ranging from 30 min to 24 h<br>Sometimes 72 h |
| - Dose | Ranging from 2 – 20 mg product/cm$^2$<br>Sometimes infinite (200 – 1200 product/cm$^2$) |
| - Species | Human and pig skin |
| - Product types | Hair dyes, preservatives, sunscreens |

15 compounds in various formulations
- Molecular weight ranges between 105 and 986 Dalton
- Lipophilicity: log Po/w ranges between -10.2 and 10.3

# Absorption under in-use conditions



# Proposed default adjustment factors for cosmetic ingredients

| $J_{max}$ (µg/cm$^2$/hr) | *Default % DA (24 h)* |
|---|---|
| Non-reactive chemicals with MW > 1000 | Negligible |
| $J_{max} < 0.1$ | 10 |
| $0.1 < J_{max} < 10$ | 40 |
| $J_{max} > 10$ | 80 |

# Worst-case assumptions in the approach

- Cosmetic ingredients are present at saturation levels
- No depletion of the ingredient occurs during the exposure period
- The formulation does not affect the skin barrier
- By using the maximal flux over the entire exposure time, the lower flux during the lag time is ignored

# Some examples

| Substance | % abs. observed | Jmax predicted | % abs. proposed default | Ratio Default/observed |
|---|---|---|---|---|
| Diethanolamine | 2.4 | 44.01 | 80 | 33.3 |
| p-phenylenediamine | 2.4 | 10.73 | 80 | 33.3 |
| Ethyl lauryl arginate HCl | 5.4 | 13.14 | 80 | 14.8 |
| Caffeine | 1.44 | 2.42 | 40 | 27.8 |

# Application of TTC to cosmetic products: a step-wise approach

Define product type, use, skin surface, concentration ingredient

Estimate external exposure

Estimate skin absorption

Apply retention factor for rinse-off products

Establish use pattern

Calculate adjusted internal exposure

Calculate total (aggregate) exposure, when relevant

Use average aggregate internal dosage in TTC decision tree

# Conclusions

The TTC concept is an appropriate tool to evaluate/prioritise the need for toxicity testing and assessment of substances present in cosmetic end products, including impurities or degradation products. The proposed concept does not apply to local effects.

Conservative default adjustment factors were established for skin absorption, to be used when experimental data are not available. Evaluation of these factors using relevant experimental data may lead to further refinement.

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