

Threshold of Toxicological Concern TTC for cosmetics & beyond

Sue Barlow - COSMOS SAB

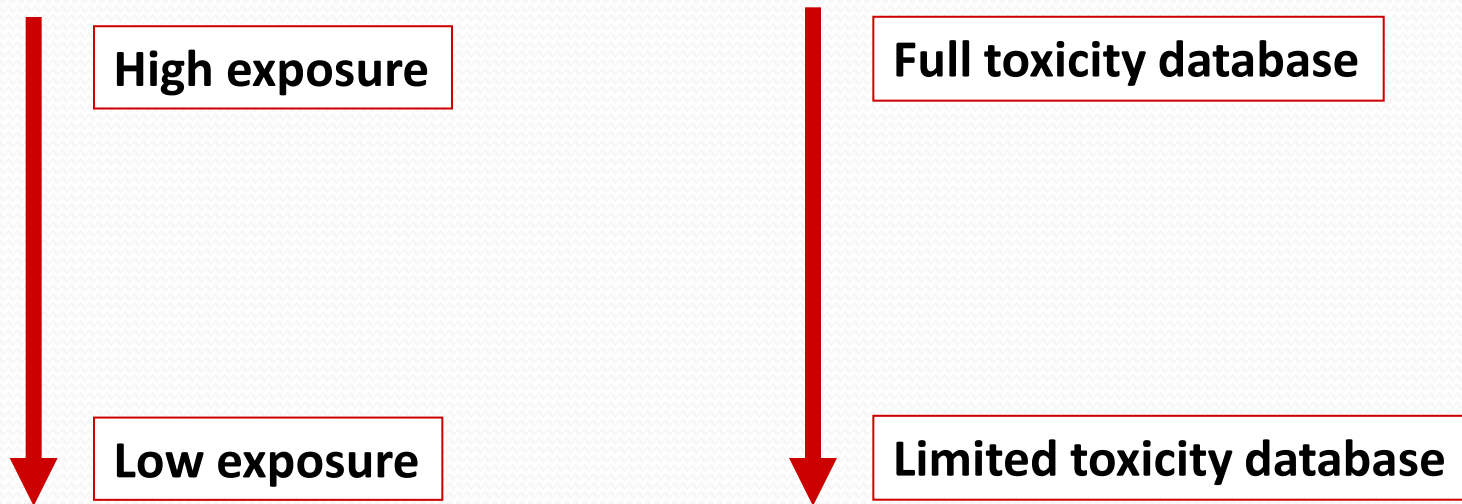
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BACKGROUND TO THE TTC CONCEPT

The amount of toxicity testing required on any chemical under evaluation is directly related to the predicted human exposure



Is there a level of exposure that is so low that we can conclude there is little concern, even in the absence of any toxicity data on that chemical?



Paracelsus 1493-1541

“All substances are
poisonous.

There is none which is
not a poison.

The right dose
differentiates a poison
from a remedy.”

WHAT IS THE TTC CONCEPT?

- Based on the fundamental principle that toxicity is related to dose and duration of exposure
- Non-cancer effects: have thresholds below which toxicity does not occur
- Cancer effects: at very low exposure levels the likelihood of tumours is zero to very small
- For chemicals of unknown toxicity, human exposure thresholds can be established, below which there would be **a low probability** of adverse effects on health

WHAT INFORMATION DO WE NEED?

- Chemical structure
- Human exposure

DEVELOPMENT OF HUMAN EXPOSURE THRESHOLD VALUES (TTC VALUES)

- Two types of TTC Value
 - Non-cancer effects
 - Cancer effects

DEVELOPMENT OF TTC VALUES FOR NON-CANCER EFFECTS

- When toxicity data are available for several members of a closely-related structural group it is possible to predict the nature of any toxicity and the potency of unstudied members using **structure-activity relationships**
- Can this approach be extended to “the world of chemicals” to identify larger groups of chemicals sharing broadly similar functional groups?
- For chemicals sharing broadly similar functional groups, can human exposure threshold values be identified below which toxicity for non-cancer effects is unlikely?

ASSIGNING CHEMICALS TO STRUCTURAL CLASSES: CRAMER DECISION TREE

Cramer, Ford & Hall 1978 classified chemicals into three structural classes based on:

- Toxicity conferred by certain structural groups
- Whether the substance occurred naturally in food
- Whether it was naturally present in the body
- What was known about its metabolism

(Food & Cosmetics Toxicology 16, 255-276, 1978)

CRAMER DECISION TREE ON STRUCTURAL CLASSES

Class I

Substances with simple structure with efficient metabolism
suggesting a low order of toxicity

Class III

Substances with structures that permit no strong initial
presumption of safety or which suggest significant toxicity

Class II

Anything that cannot be put into Class I or Class III

Cramer et al. 1978 separated chemicals into 3 structural classes via a series of questions I = low, II = medium, III = high toxicity

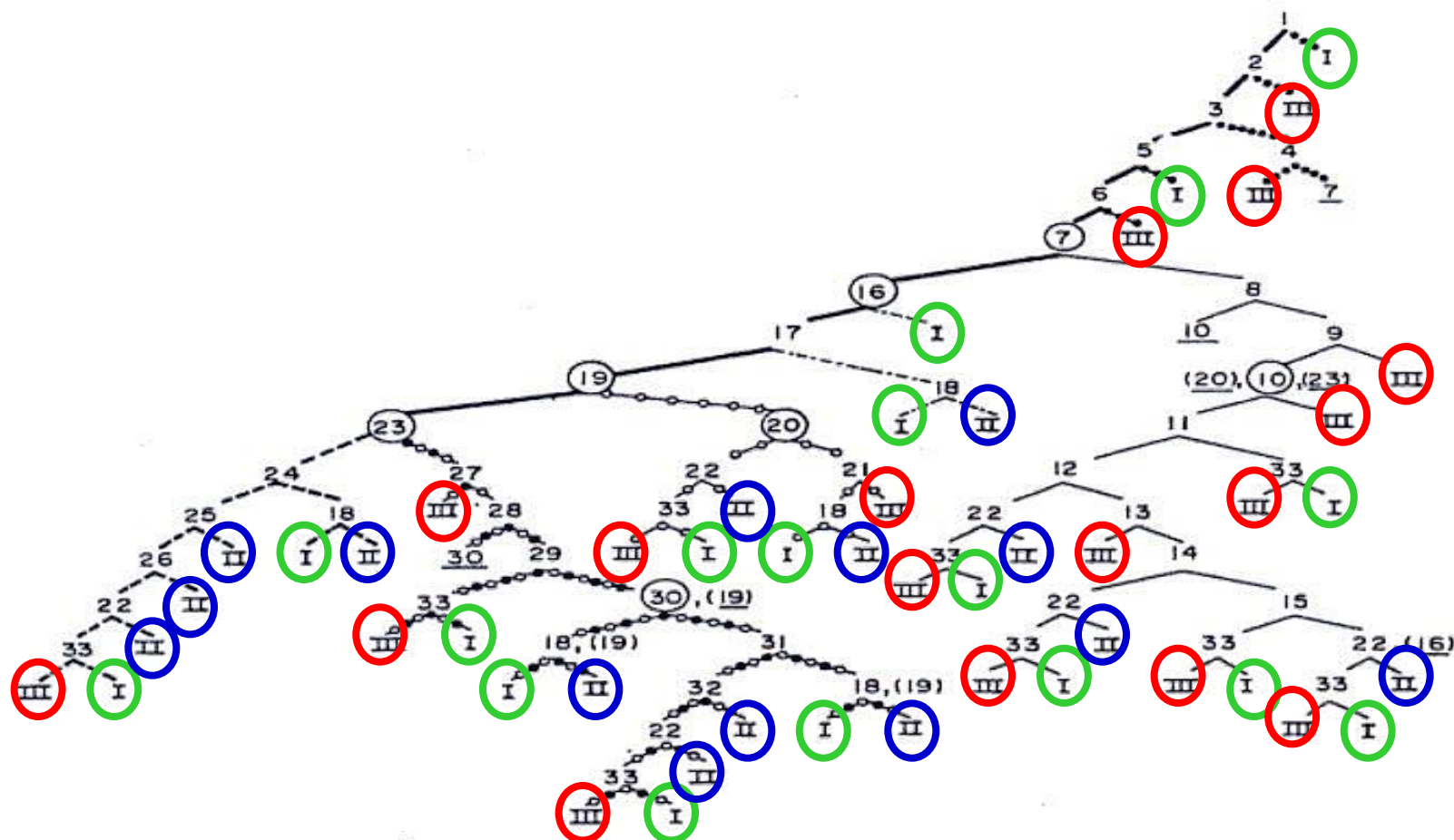


Fig. 1. A schematic diagram of a decision tree for the estimation of probable toxicity. Assessors should (a) start with question 1, (b) proceed by 'no' ✓ or 'yes', (c) move from any underscored number encountered to same circled number and (d) proceed to final classes I, II or III. Working downwards through the tree, the symbols designate the following groupings: biological normality (●●●); high and low toxicity (●—●); heterocyclics (—); terpenoids (—·—); aliphatics (—○—○—); aromatics (—○—●—○—).

DEVELOPMENT OF THRESHOLD VALUES from non-cancer data

Munro et al. 1996

- Compiled a database of existing **oral** toxicity data on 613 substances (sub-chronic, chronic, reproductive, developmental toxicity)
- Plotted the distribution of their toxic potencies, expressed as their no-observed-effect levels (NOELs), with substances grouped according to the Cramer structural classification scheme

(Food & Chemical Toxicology 34, 829-867, 1996)

DEVELOPMENT OF THRESHOLD VALUES

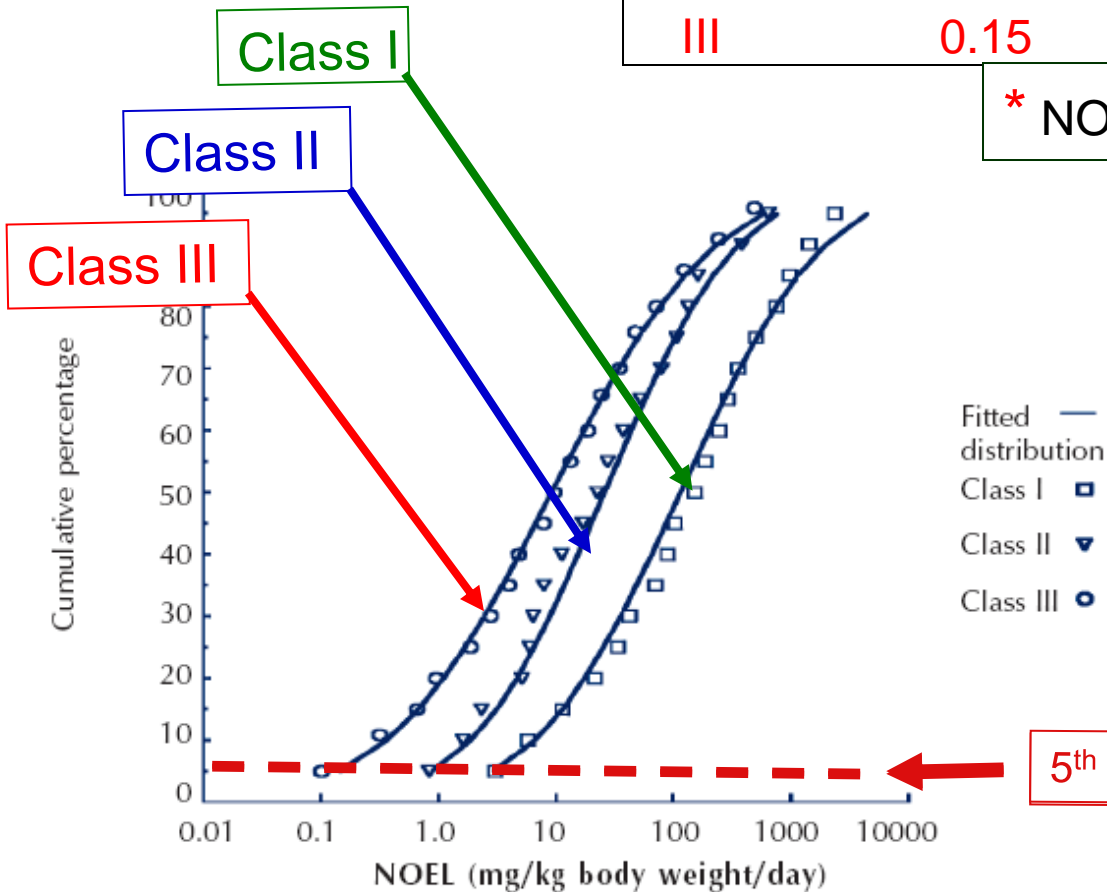
from non-cancer data

- Used 5th percentile NOEL values from the distributions for each of the three Cramer structural groups
- Divided the 5th percentile NOEL values by a factor of 100 to take account of uncertainties
- Multiplied the resulting amounts by 60 to represent a 60kg adult
- Obtaining human exposure threshold values expressed as micrograms/person/day

Munro et al. 1996

Class	5%ile NOEL (mg/kg/day)	Human threshold (μ g/day) *
I	3.0	1800
II	0.91	540
III	0.15	90

* NOEL/100 X 60kg bwt



Reprinted from *Food and Chemical Toxicology* Vol 34, Munro IC, Ford RA, Kennepohl E and Sprenger JG; Correlation of a structural class with no-observed-effect levels: a proposal for establishing a threshold of concern, pp 829-867, Copyright 1996, with permission from Elsevier.

Probabilities not certainties

If human exposure is below the threshold, the substance can be judged, with reasonable confidence to present a low probability of risk

(Munro et al. Food & Chemical Toxicology 34, 829-867, 1996)



DEVELOPMENT OF THRESHOLD VALUES from cancer data

What if untested substances have
genotoxic or carcinogenic
properties?

US THRESHOLD OF REGULATION POLICY

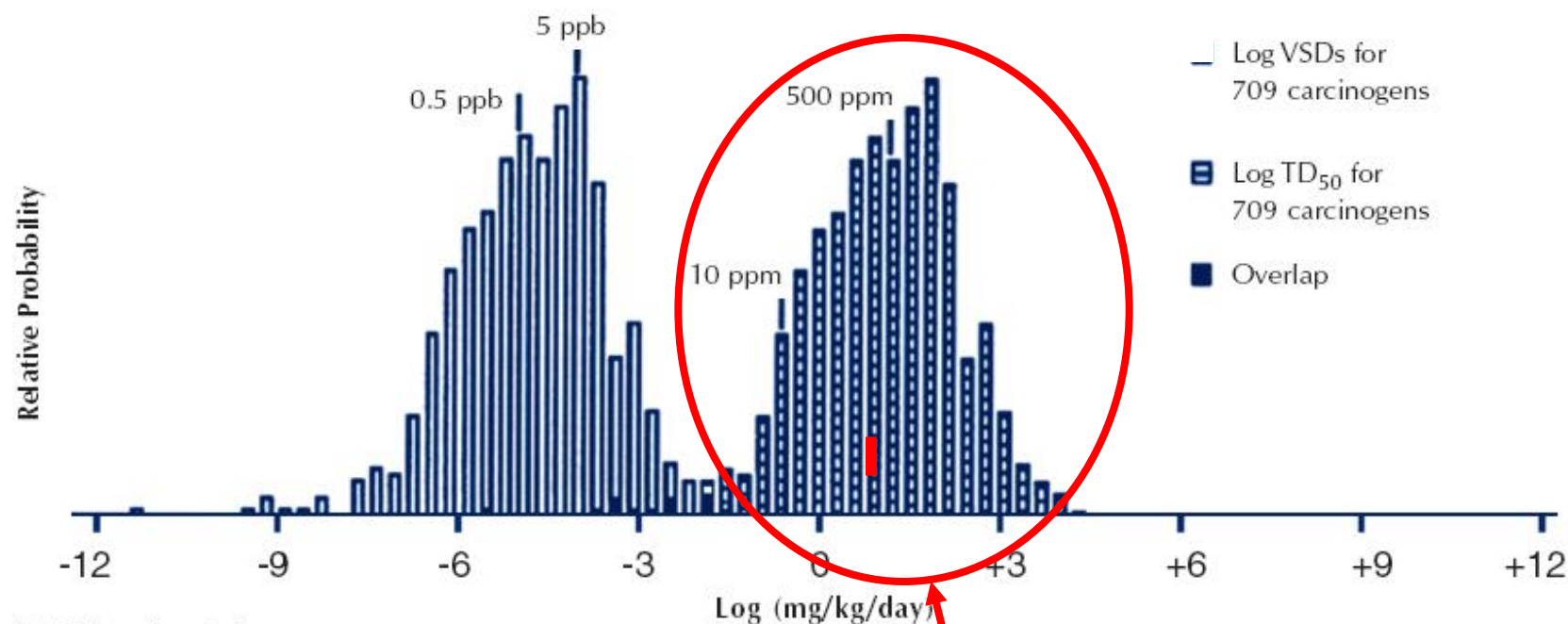
- FDA derived a 'Threshold of Regulation' (TOR) for FCMs based on data from the Carcinogenic Potency Data Base (Gold et al. - now has >700 substances)
- For each chemical, calculated the TD_{50} for most sensitive species and sex (TD_{50} = lifetime daily dose inducing tumours in 50% of the animals, corrected for background tumours in controls)
- Used linear extrapolation from plot of TD_{50} s to derive upper bound values for oral doses assumed to cause 1 in a million lifetime increase in risk of cancer ('virtually safe doses' VSDs)
- Selected 0.5 ppb as threshold value, equivalent to 1.5 $\mu\text{g}/\text{person}/\text{day}$ assuming adults consume 1.5 kg food and 1.5L of fluids daily

TOR policy 1995: Any substance originating from FCM present in food at a concentration below 0.5 ppb is exempt from regulation (and testing)

RODENT CARCINOGENICITY DATA BASE

FIGURE 1

Distribution of TD_{50} s for chemical carcinogens and extrapolation to a 1 in a million risk



VSD: Virtually safe dose

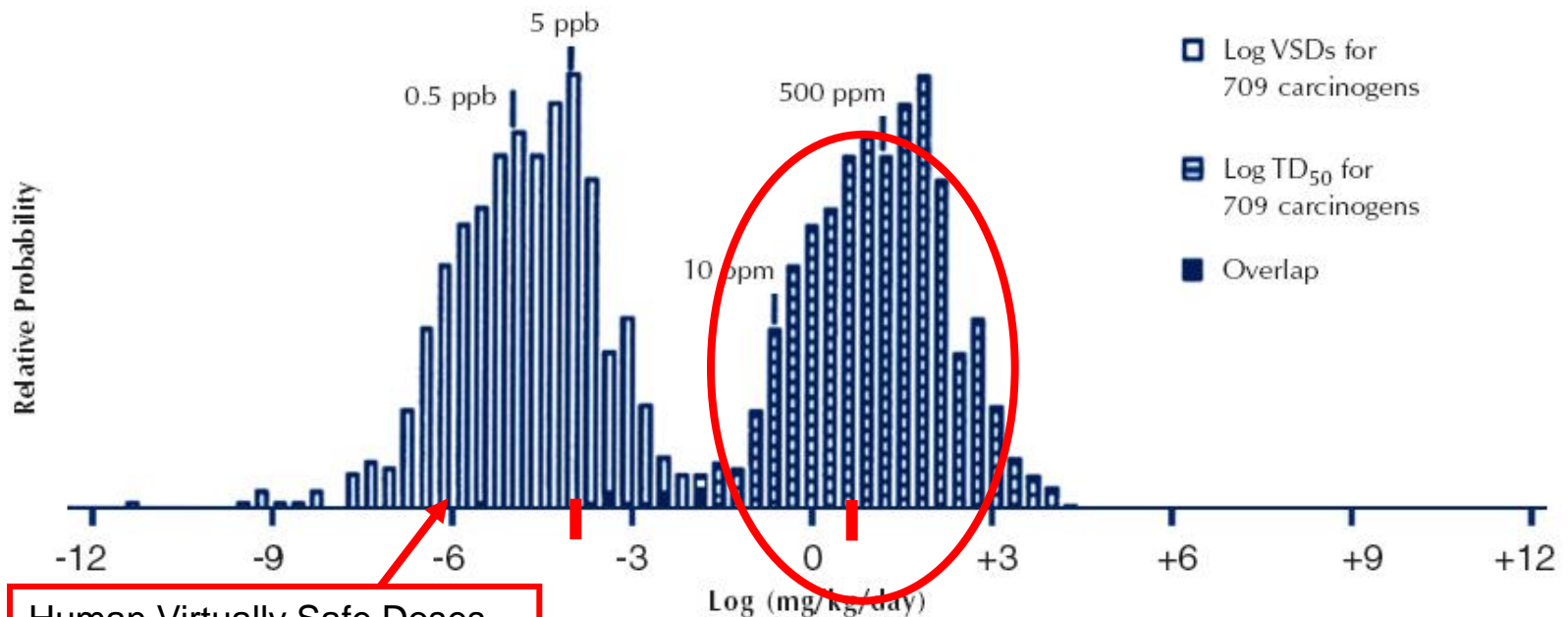
Reprinted from *Food and Chemical Toxicology* Vol 37. Cheeseman MA, Machuga EJ and Bailey AB; A tiered approach to threshold of regulation, pp387-412, Copyright 1999, with permission from Elsevier.

Rodent TD_{50}

RODENT CARCINOGENICITY DATA BASE

FIGURE 1

Distribution of TD_{50} s for chemical carcinogens and extrapolation to a 1 in a million risk

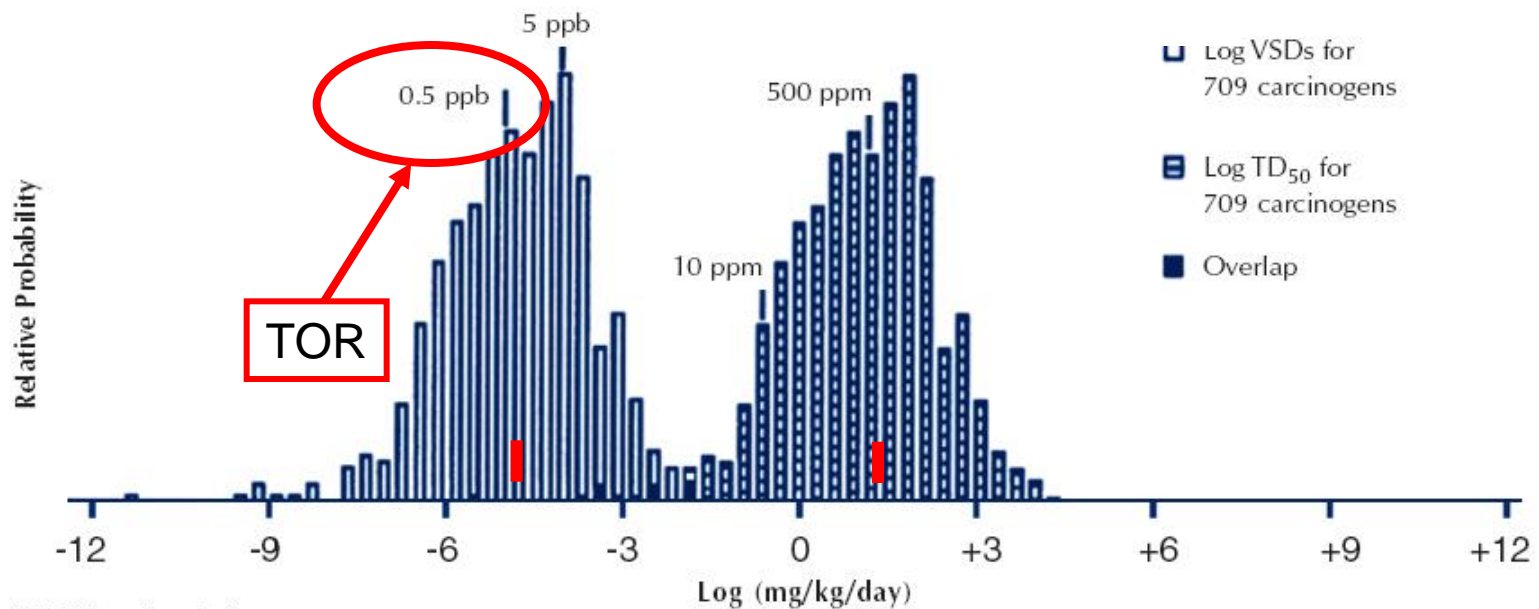


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RODENT CARCINOGENICITY DATA BASE

FIGURE 1

Distribution of TD_{50} s for chemical carcinogens and extrapolation to a 1 in a million risk



VSD: Virtually safe dose

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IS THE TOR VALUE SUFFICIENTLY PROTECTIVE?

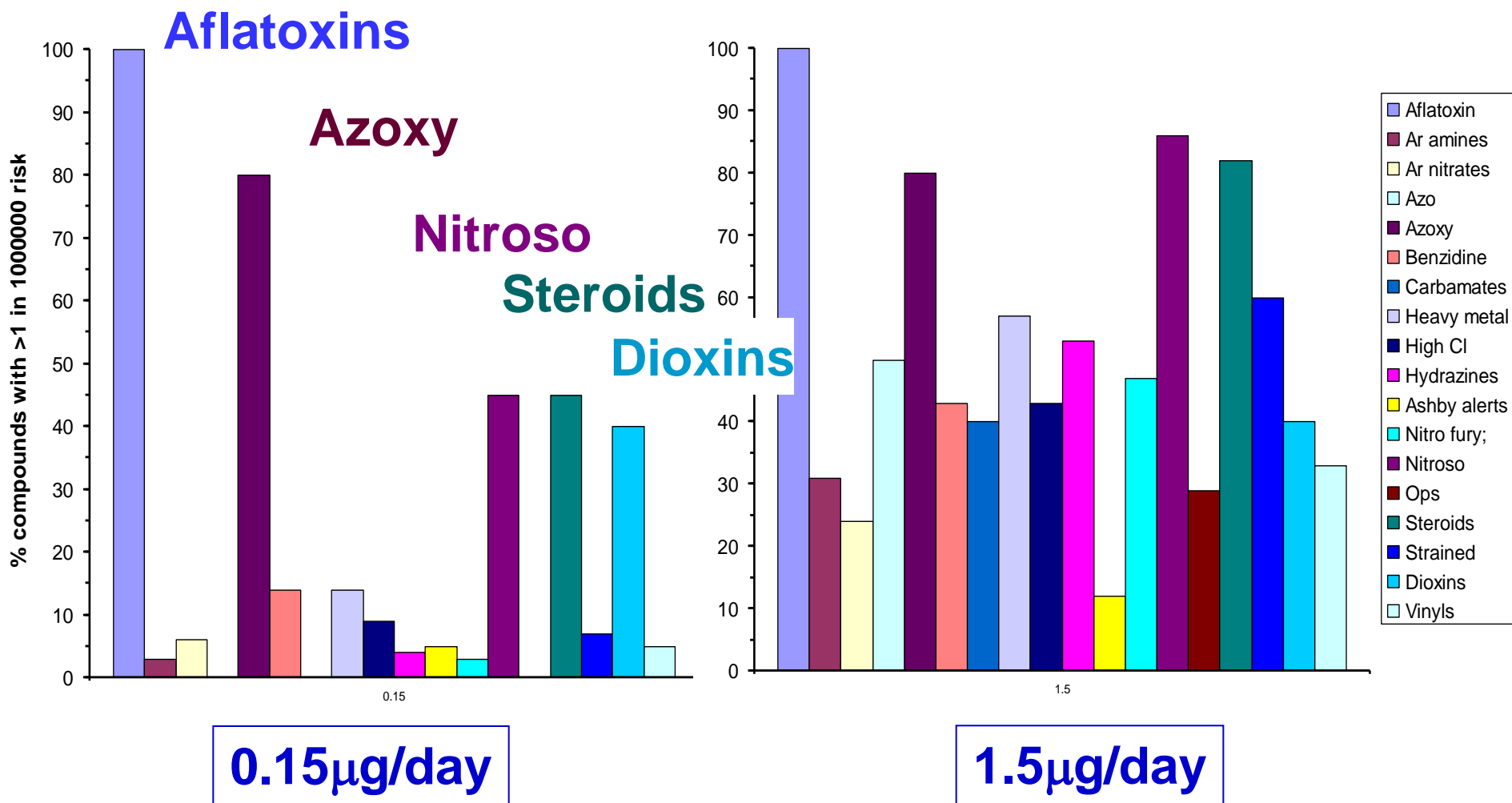
- It is evident that at the TOR value of 1.5 µg/person/day a number of carcinogens have potencies that could give rise to an estimated risk of greater than 1 in a million
- So subsequent work aimed to define a more conservative threshold and to identify high potency carcinogens that could be excluded from the TTC approach

EXCLUSION OF HIGH POTENCY CARCINOGENS

Work of Cheeseman et al. (1999) and Kroes et al. (2004)

- At a lower threshold value of $0.15 \mu\text{g}/\text{person}/\text{day}$, a much greater proportion of carcinogens have an estimated risk of less than 1 in a million compared with the TOR value of $1.5 \mu\text{g}/\text{person}/\text{day}$
- In addition, some more potent carcinogens, termed the 'cohort of concern' could be excluded from the TTC approach

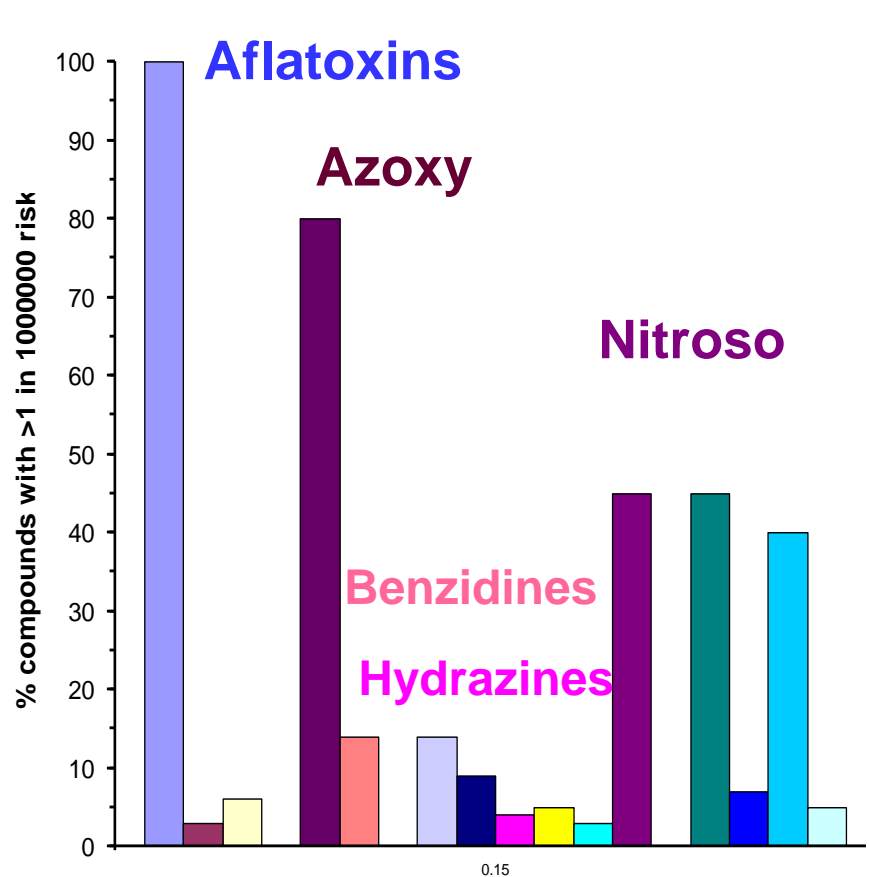
% Compounds in the group with a calculated risk >1 in 1,000,000 at different levels of daily intake



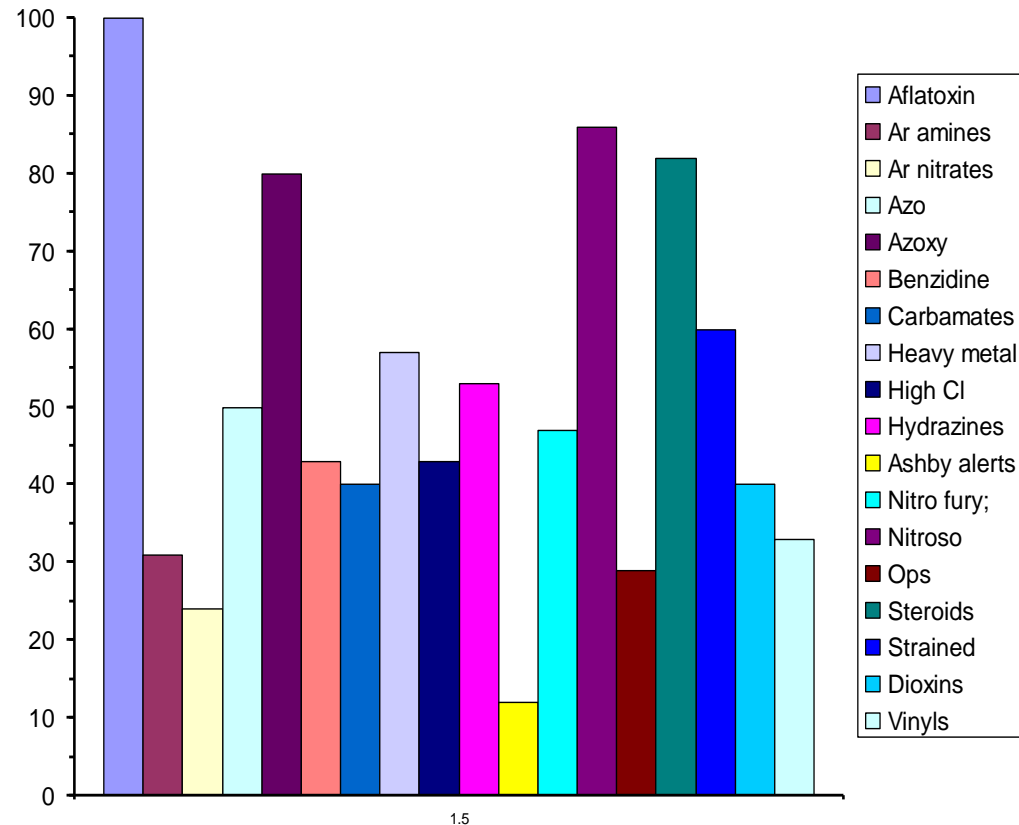
Remove compounds for which there would be a threshold

Steroids Dioxins

% Compounds in the group with a calculated risk > 1 in 1,000,000 at different levels of daily intake



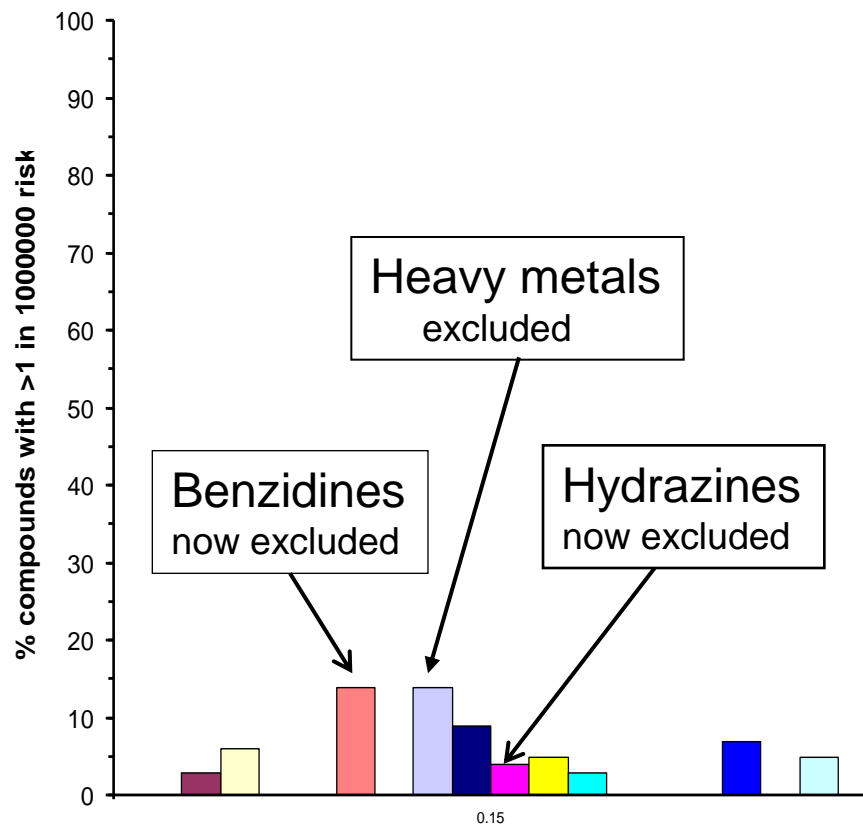
0.15 $\mu\text{g}/\text{day}$



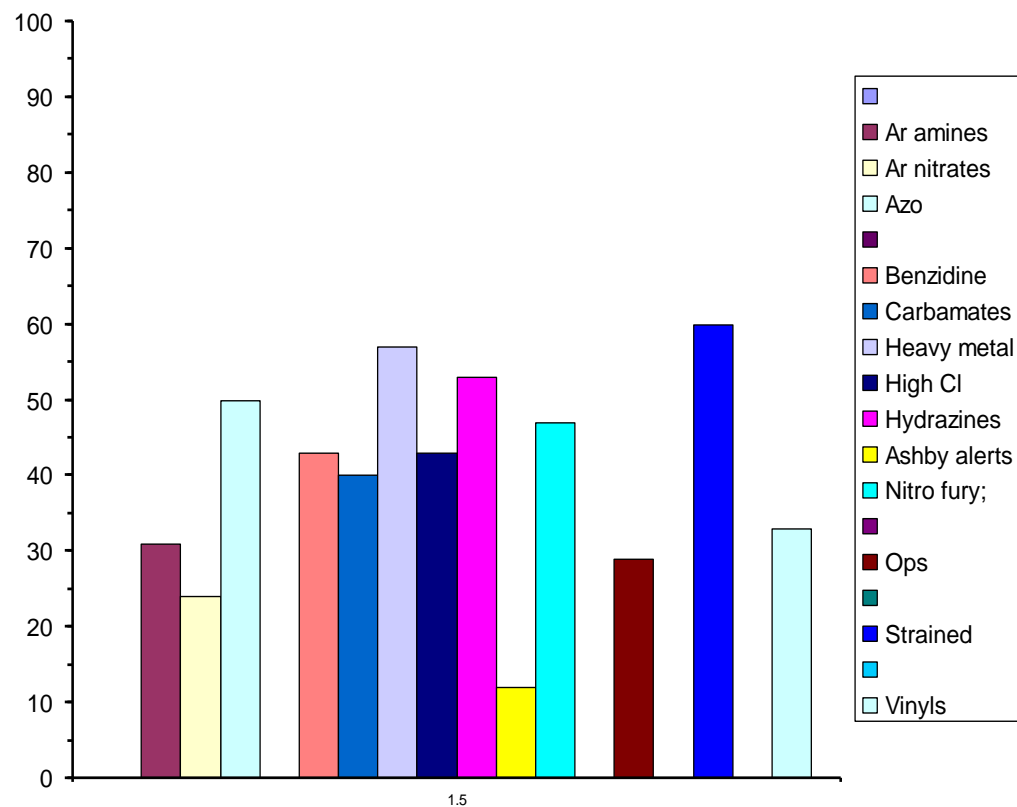
1.5 $\mu\text{g}/\text{day}$

Remove compounds which are the most potent

% Compounds in the group with a calculated risk > 1 in 1,000,000 at different levels of daily intake



0.15µg/day



1.5µg/day

For the remaining compounds here would be a high probability that any cancer risk would be <1 in a million at an intake of 0.15 µg/day

ESTIMATING HUMAN EXPOSURE

**Reliable exposure estimates are key for TTC
i.e. they should not underestimate exposure**

- If have good exposure data, use it
- If not, estimate worst-case exposure
- In the case of cosmetics, exposure can be dermal, oral or inhalational
- Translate dermal exposures to oral/systemic exposures using models for skin permeability (COSMOS)

IS THE CRAMER DECISION TREE DIFFICULT TO USE ?

- Toxtree software is available

<http://sourceforge.net/projects/toxtree/>

- It allows a drawn chemical structure to be imported, or can use chemical name, CAS No or SMILES code
- It takes the structure sequentially through the questions until it gives an answer that allows the structure to be classified in either Cramer Class I, Class II or Class III

APPLYING QUESTIONS TO A QUERY SUBSTANCE

Rules

Rules

- Q1.Normal constituent of the body
- Q2.Contains functional groups associated with enhanced toxicity
- Q3.Contains elements other than C,H,O,N,divalent S
- Q4.Elements not listed in Q3 occurs only as a Na,K,Ca,Mg,N salt, sulphamate, sulph...
- Q5.Simply branched aliphatic hydrocarbon or a common carbohydrate
- Q6.Benzene derivative with certain substituents
- Q7.Heterocyclic
- Q8.Lactone or cyclic diester
- Q9.Lactone, fused to another ring, or 5- or 6-membered a,b-unsaturated lactone?
- Q10.3-membered heterocycle
- Q11.Has a heterocyclic ring with complex substituents.
- Q12.Heteroaromatic
- Q13.Does the ring bear any substituents?
- Q14.More than one aromatic ring
- Q15.Readily hydrolysed
- Q16.Common terpene
- Q17.Readily hydrolysed to a common terpene
- Q18.One of the list (see explanation)
- Q19.Open chain
- Q20.Aliphatic with some functional groups (see explanation)
- Q21.3 or more different functional groups
- Q22.Common component of food
- Q23.Aromatic
- Q24.Monocarbocyclic with simple substituents
- Q25.Cyclopropane, etc. (see explanation)
- Q26.Monocycloalkanone or a bicyclic compound
- Q27.Rings with substituents
- Q28.More than one aromatic ring
- Q29.Readily hydrolysed
- Q30.Aromatic Ring with complex substituents
- Q31.Is the substance an acyclic acetal or ester of substances defined in Q30?
- Q32.Contains only the functional groups listed in Q30 or Q31 and those listed below.
- Q33.Has sufficient number of sulphonate or sulphamate groups

1. Normal constituent of the body?

Decision node

Decision node: Q1.Normal constituent of the body

If 'NO' go to: Q2.Contains functional groups associated with enhanced toxicity

If 'YES' assign: Low (Class I)

Rule ID: 1 Rule title: Normal constituent of the body

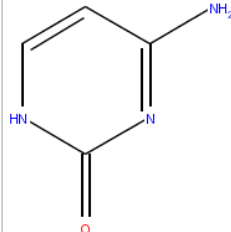
Rule explanation:

Is the substance a normal constituent of the body, or an optical isomer of such?

This question throws into class I all normal constituents of body tissues and fluids, including normal metabolites. Hormones are excluded, as are, by implication, the metabolites of environmental and food contaminants or those resulting from disease state.

Note the answer of the question relies on an incomplete list of compounds, identified by an expert as a normal body constituents. If you believe a query compound is wrongly identified as a such, or not recognised, please consult and/or update the list. C:\Ideas\consult\toxTree-v1.00\toxTree\bodymol.slf

Example with answer 'YES'



There are example molecules for each rule outcome. Select which one to display.

☒ Yes branch ☐ No branch

Yes - cytosine: Class I (low concern)
No proceed down the tree (Q2)

Slide from Andrew Worth JRC

APPLYING QUESTIONS TO A QUERY SUBSTANCE

Rules

Rules

- Q1.Normal constituent of the body
- Q2.Contains functional groups associated with enhanced toxicity
- Q3.Contains elements other than C,H,O,N,divalent S
- Q4.Elements not listed in Q3 occurs only as a Na,K,Ca,Mg,N salt, sulphamate, sulph...
- Q5.Simply branched aliphatic hydrocarbon or a common carbohydrate
- Q6.Benzene derivative with certain substituents
- Q7.Heterocyclic
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- Q9.Lactone, fused to another ring, or 5- or 6-membered α,β -unsaturated lactone?
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- Q15.Readily hydrolysed**
- Q16.Common terpene
- Q17.Readily hydrolysed to a common terpene
- Q18.One of the list (see explanation)
- Q19.Open chain
- Q20.Aliphatic with some functional groups (see explanation)
- Q21.3 or more different functional groups
- Q22.Common component of food**
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- Q30.Aromatic Ring with complex substituents
- Q31.Is the substance an acyclic acetal or ester of substances defined in Q30?
- Q32.Contains only the functional groups listed in Q30 or Q31 and those listed below.
- Q33.Has sufficient number of sulphonate or sulphamate groups

Decision node

Decision node: Q22.Common component of food

If 'NO' go to: Q33.Has sufficient number of sulphonate or sulphamate groups

If 'YES' assign: Intermediate (Class II)

Rule ID: 22 Rule title: Common component of food

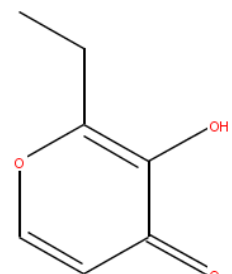
Rule explanation:

Is the substance a *common component of food* (C) or *structurally closed related* to a common component of food?

(C) Common component of food. In something as diverse, changing and occasionally uncertain as natural occurrence, it is only possible to define a guideline, not a firm rule. For a decision tree, the term common component of food denotes a substance that has been reported in the recognised literature as occurring in significant quantity (approximately 50 ppm or more) in at least one major food, or in trace quantities at the ppm level or less in several foods, including minor or less frequently consumed foods. The latter include spices, herbs and ethnic specialities. This definition excludes natural or man made contaminants and hormones.

Note the answer of the question relies on an incomplete list of compounds, identified by an expert as a common component of food. If you believe a query compound is wrongly identified as a such, or not recognised, please consult and/or update the list. *C:\ideaconsult\toxTree-v1.0\toxTree\foodmol.sdf*

Example with answer 'YES'



There are example molecules for each rule outcome. Select which one to display.

☒ Yes branch ☐ No branch

22. Common component of food?

Yes - ethyl maltol (flavour): Class II (intermediate class)

No proceed down the tree

TOXTREE MAIN SCREEN: EXAMPLE VINCLOZOLIN

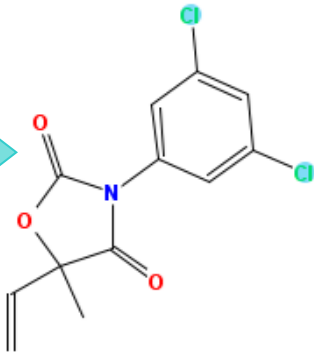
Toxtree (Estimation of Toxic Hazard - A Decision Tree Approach) v2.6.0

File Edit Chemical Compounds Toxic Hazard Method Help

Chemical identifier vinclozolin Go!

Available structure attributes	
Cramer rules	High (Class III)
Names	vinclozolin
cdk:Comment	Retrieved from http://ap...
http://www.opentox.org...	50471-44-8
http://www.opentox.org...	N-3,5-dichlorophenyl-5-...
http://www.opentox.org...	256-599-6
http://www.opentox.org...	3-(3,5-dichlorophenyl)-5-...
http://www.opentox.org...	F5CWZHGZWWDELK-LBP...
http://www.opentox.org...	InChI=1S/C12H9Cl2NO3...
http://www.opentox.org...	30.11.2010
http://www.opentox.org...	CC1(OC(=O)N(C1=O)c2...

Structure diagram



First Prev 1 / 1 Next Last

Completed.

Toxic Hazard by Cramer rules

Estimate

Low (Class I)

Intermediate (Class II)

High (Class III)

☒ Verbose explanation

Cramer rules

- Q1. Normal constituent of the body **No** vinclozolin
- Q2. Contains functional groups associated with enhanced toxicity **No** vinclozolin
- Q3. Contains elements other than C,H,O,N,divalent S **Yes** vinclozolin
- Q4. Elements not listed in Q3 occurs only as a Na,K,Ca,Mg,N salt, sulphamate, sulphonate, sulphate, hydrochloride ... **No** Class **High (Class III)** vinclozolin

Compound properties

Prediction

Compound structure

Reasoning

Slide from
Andrew Worth
JRC

WHAT IS COSMOS DOING ON TTC?

- Developed a COSMOS TTC database containing:
 - 560 chemicals used in cosmetic products (from US and EU inventories, plus cosmetic-use chemicals from Munro DB)
 - Repeat-dose oral toxicity data on non-cancer effects and the lowest NOAEL for each chemical
- QC work: Chemicals within lowest 10th percentile of NOAEL distribution for each Cramer classes, or with differing NOAELs
- TTC values for chemicals used in cosmetics will be determined for each Cramer class
- Also under development
 - Separate database on oral and dermal absorption data
 - Decision framework for oral-to dermal extrapolation

SUMMARY OF TTC VALUES

as proposed by Munro et al. 1996

Chemical structure	TTC Value $\mu\text{g}/\text{person}/\text{day}$	TTC value $\mu\text{g}/\text{kg bw}/\text{day}$
Genotoxicity SA	0.15	0.0025
OPs & carbamates	18	0.3
Cramer Class III	90	1.5
Cramer Class II	540	9
Cramer Class I	1800	30

Does the substance have a known structure and are exposure data available?

No

TTC approach cannot be applied

Yes

EFSA GENERIC SCHEME

Is the substance a member of an exclusion category? *

Yes

No

Is there a structural alert for genotoxicity (including metabolites)?

Yes

Exposure > 0.0025 µg/kg bw/day?

Yes

No

Low probability of health effect **

Substance requires non-TTC approach (toxicity data, read-across, etc)

Low probability of health effect **

No

No

Exposure > 0.3 µg/kg bw/day? *** OP/Carbamate TTC

Yes

Is substance an OP/Carbamate?

Yes

No

No

Exposure > 1.5 µg/kg bw/day? *** Cramer Class III TTC

Yes

Is substance in Cramer Class II or III?

Yes

No

No

Exposure > 30 µg/kg bw/day? *** Cramer Class I TTC

Yes

* Exclusion categories

High potency carcinogens; Inorganic substances; Metals and organometallics; Proteins; Steroids; Substances known/predicted to bioaccumulate; Nanomaterials; Radioactive substances; Mixtures.

** If exposure of infants < 6 months is in range of TTC
→ consider if TTC is applicable

*** If exposure only short duration
→ consider margin between human exposure & TTC value

SUMMARY & CONCLUSIONS

- In the absence of toxicity data, the TTC approach is sufficiently robust to be used to screen substances with low human exposures
- Oral TTC values proposed by Munro since confirmed by others to be conservative
 - COSMOS TTC database will propose TTC values for cosmetics
 - Development of dermal and inhalational TTC values ongoing
- N.B. It is a probability-based tool
 - Exposure to a substance below the relevant TTC value may still pose a potential risk, with a probability estimated to lie between zero and 5%

Thank you!

