How Toxicity Data are Used in the Process of Hazard Identification and Dose-Response Assessment

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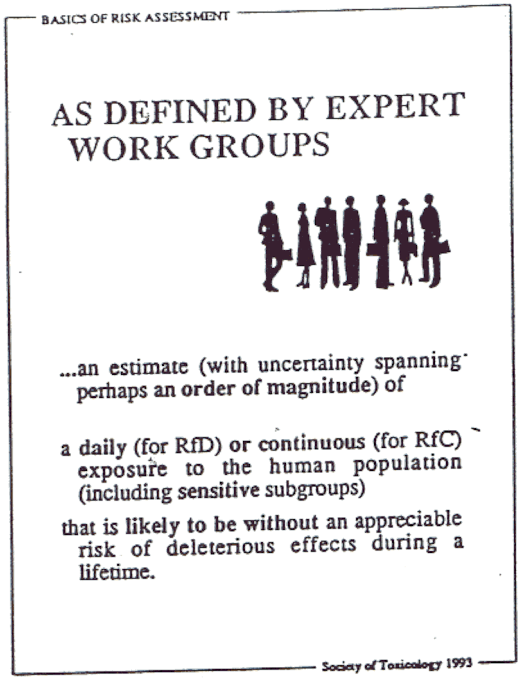
DISCUSSION POINTS:

The purpose of this lecture is to inform you of basic principles in hazard identification and dose response assessment.

It is a common (although not exclusive) practice in risk assessment to distinguish between cancer and non cancer health endpoints.

Thus, this lecture will be divided into these two areas.



DISCUSSION POINTS:

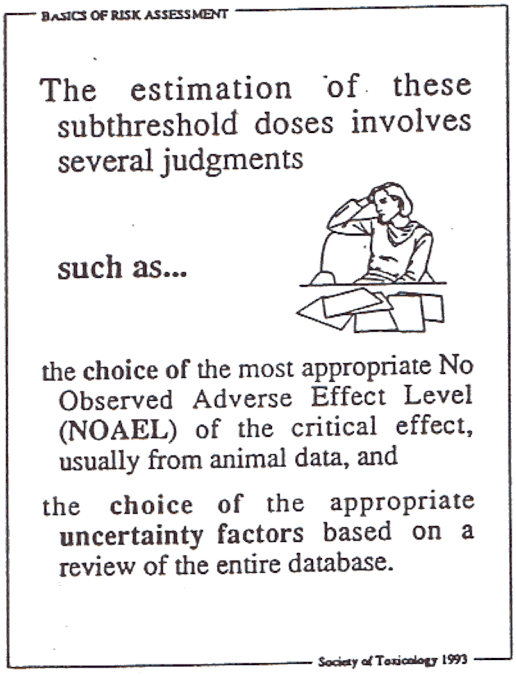
PART 1: NONCANCER RISK ASSESSMENT

Expert work groups throughout the world support the use of a "safe" dose concept in noncancer risk assessment, and define terms and conditions for its use.

This safe or subthreshold dose often goes by different names, such as WHO's Tolerable Intake (TI); U.S. ATSDR's Minimum Risk Level (MRL); U.S. FDA's Acceptable Daily Intake (ADI).; and U.S. EPA's Reference Dose (RfD) or Reference Concentration (RfC).

For example, the U.S. EPA uses the terms Reference Dose (RfD) for oral and Reference Concentration (RfC) for inhalation exposures. These terms are defined on the adjacent slide.

Many of the underlying assumptions, judgements on critical effect, and choices of uncertainty factors (or safety factors) to estimate these various subthreshold doses are similar.



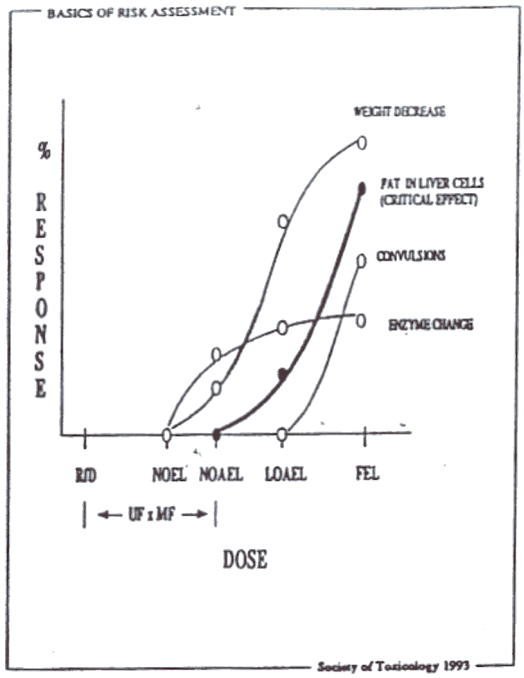
DISCUSSION POINTS:

A number of judgements are used in the determination of these subthreshold doses.

Two large ones are ...

what constitutes an adverse effect or syndrome of adverse effects with a given chemical, and then what is the critical effect, and

what is the appropriate composite uncertainty (or safety) factor to use based on considerations of the entire toxicity data base.



DISCUSSION POINTS:

All toxcity data are reviewed in order to determine the critical effect.

The critical effect is the first adverse effect(s) or its know precursor(s) that occurs as dose rate increases.

Some effects are clearly not adverse, nor precursors; other are clearly adverse or precursors; many effects are of uncertain adversity.

The judgement of what effects are adverse is often difficult and often depends on the spectrum of effects seen in the chosen study or other studies in the data base.

For definitions of the EPA terms used on the adjacent slide, please see a separate handout. Also note that health organizations throughout the world use similar terms. This listing is only one example of how to define the concept of critical effect.

NOEL = No Observed Effect Level

NOAEL = No Observed Adverse Effect Level

LOAEL = Lowest Observed Adverse Effect Level

FEL = Frank Effect Level

UF = Uncertainty Factor, MF = Modifying Factor

BASICS OF RISK ASSESSMENT

UNCERTAINTY FACTORS

FACTOR*		EXTRAPOLATION
H	10	AVERAGE HUMAN TO SENSITIVE HUMAN
A	10 or 3**	ANIMAL TO HUMAN
S	≤10	SHORT TERM TO LONG TERM EXPOSURE
L	≤10	LOAEL TO NOAEL
D	≤10	MINIMUM TO COMPLETE DATA BASE

*These factors are as used by the U.S. EPA. Other health organiztions use similar factors. In EPA, The maximum UF for any given data base is 10,000. Data bases weaker than this are judged too uncertain to estimate RfD/RfCs.

**For RfCs the default UF for area "A," animal to human, is 3.

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DISCUSSION POINTS:

Uncertainty Factors are necessary reductions in the dose to account for missing data.

Health organizations throughout the world treat these areas of uncertainty somewhat differently. Some, like the U.S.EPA on the slide, call out explicit areas.

So what is ideal data to determine a protective value? When would an uncertainty factor not be needed?

BASICS OF RISK ASSESSMENT

UF PRACTICE (H x A x S x L x D)

Any one area, generally 10

Any two areas, generally 100

Any three areas, generally 1000

Any four areas, generally 3000

All five areas, generally 10,000

Intermediate factors are generally developed on a logarithmic basis.

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DISCUSSION POINTS:

The composite UF is a judgement regarding the strengths and weaknesses of the entire data base.

As UFs increase in number, the composite "protectiveness" of resulting value (e.g., ADI) generally increases. This implies that as more data are brought into the data base for the determination of the ADI, fewer uncertainty factors would be needed and the value of the ADI should increase. This is generally what occurs.

Most agencies that estimate these values recognize this composite "protectiveness" and roll several areas of uncertainty together.

BASICS OF RISK ASSESSMENT

Accuracy of an ADI

ADIs, MRLs, RfDs, RfCs or TIs are considered to be accurate estimates of doses **below a toxicity threshold,** because they are based on a review of all toxicity data and individual uncertainty factors are considered to be somewhat protective. The use of several UFs can yield conservative estimates.

If you play golf, accurate drives are generally those that land in the fairway, although they may be scattered over a wide area.

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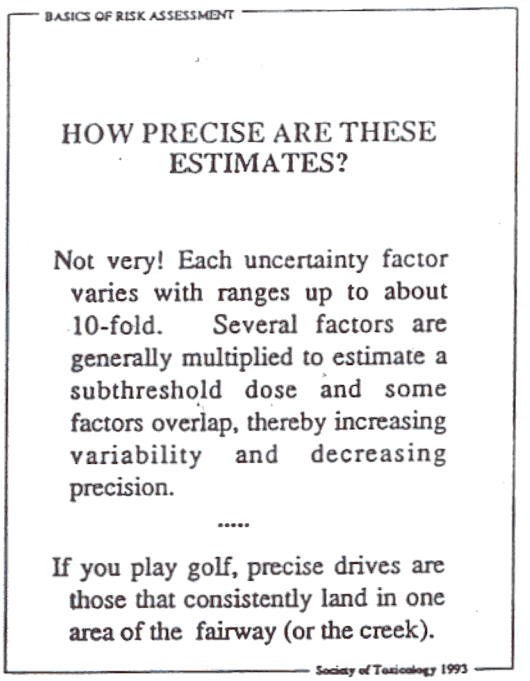
DISCUSSION POINTS:

These estimates of subthreshold dose rates are considered accurate.

However, the degree to which these dose rates are below the population threshold is not generally known.

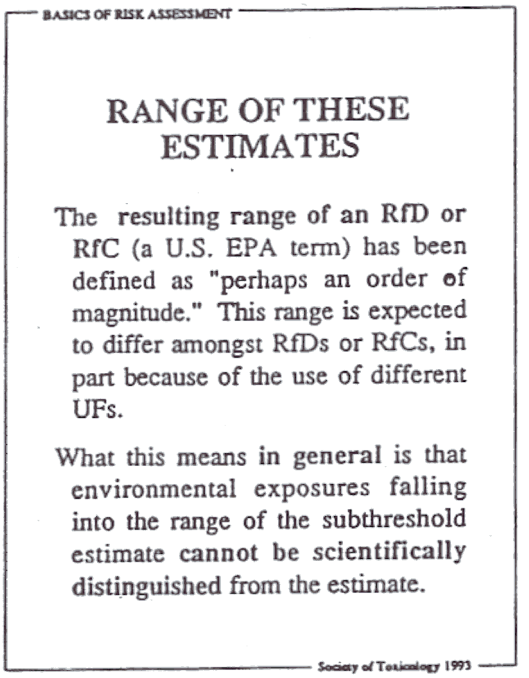
Two examples of where such estimates were not below thresholds were colbalt in beer and the sedative thalidomide. In the former case, a population sensitive to the cardiomyopathy of colbalt appeared to have been missed (thus the uncertainty factor of 10 may not have been enough). In the latter case, the critical effect, developmental toxicity, was apparently not evident in the animal toxicity data base.



DISCUSSION POINTS:

Subthreshold doses can only be as precise as their least precise component. In the case of most of these estimates, the least precise component is the uncertainty factor.

As the composite uncertainty factor grows larger, the imprecision of the resulting subthreshold doses also grows larger.

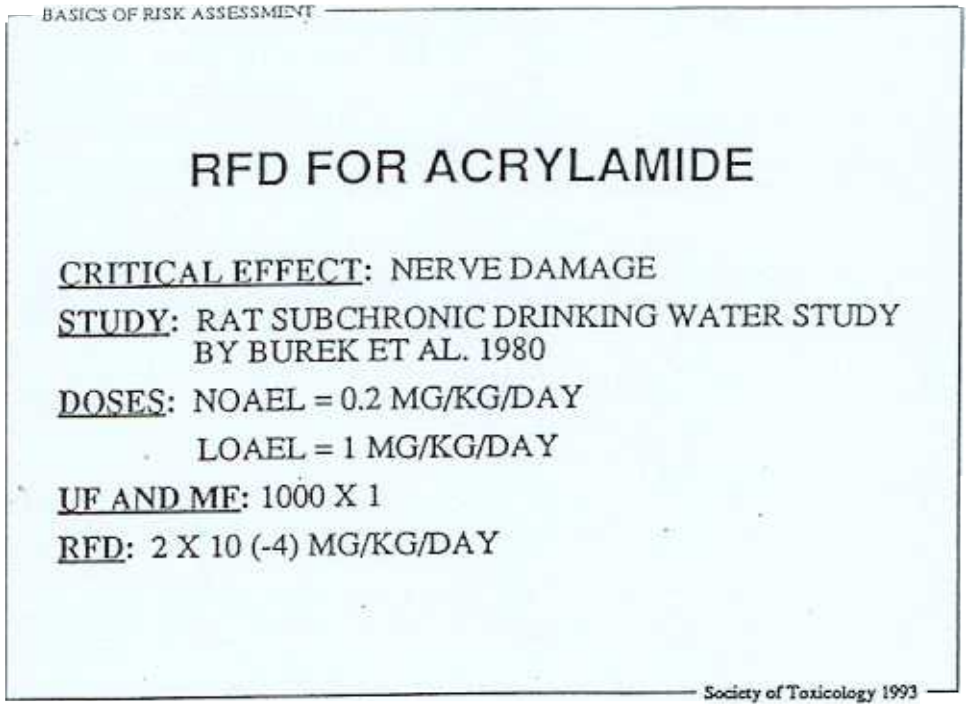


DISCUSSION POINTS:

Thus, as a result of this accuracy and precision, these subthreshold doses have an associated range.

For example, an RfD on EPA's Integrated Risk Information System (IRIS) of 1 mg/kg/day might have a range of 0.3 to 3 mg/kg/day, indicating a one-half order of magnitude both above and below the RfD.

This range might be larger or smaller depending how precise the NOAEL is and how many uncertainty factors are used to determine the subthreshold dose.



APPENDIX:

The following example on acrylamide is taken from U.S. EPA's Integrated Risk Information System (IRIS). This example shows summary information on an RfD. Also attached are definitions of some terms and additional readings. These are only a subset of terms and possible readings available for this area.

DISCUSSION POINTS:

The critical effect is considered to be nerve damage.

The left sciatic nerve was examined under light and electron microscope. No significant adverse effects were seen in the 0.05 or 0.2 groups, while the 5 and 20 mg/kg/d groups had frank nerve degeneration. In the 1 mg/kg/d group a slight, but significant, increase in peripheral axolemnal invaginations were observed under EM but not when using light microscopy.

Thus, the NOAEL was judged to be 0.2 mg/kg/day and the LOAEL was judged to be 1 mg/kg/day.

BASICS OF RISK ASSESSMENT

ACRYLAMIDE RFD

UNCERTAINTY AND MODIFYING FACTORS

UF = 1000. THE UF OF 1000 ALOWS FOR UNCERTAINTY IN THE EXTRAPOLATION OF DOSE LEVELS FROM LABORATORY ANIMALS TO HUMANS, UNCERTAINTY IN THE THRESHOLD FOR SENSITIVE HUMANS, AND UNCERTAINTY IN THE EFFECT OF DURATION WHEN EXTRAPOLATING FROM SUBCHRONIC TO CHRONIC EXPOSURE. THE CHRONIC STUDY DID NOT ADEQUATELY ADDRESS THE LATTER UNCERTAINTY BECAUSE OF THE LACK OF A SENSITIVE MEASUREMENT OF THE CRITICAL EFFECT.

MF = 1.

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BASICS OF RISK ASSESSMENT

STUDY: HIGH

DATA BASE: MEDIUM

RFD: MEDIUM

CONFIDENCE IN THE ACRYLAMIDE RFD

THE PRINCIPAL STUDY WAS VERY WELL-DESIGNED FOR THE EVALUATION OF A LARGE NUMBER OF ENDPOINTS AND EMPLOYED A SENSITIVE MEASURE OF THE MOST APPROPRIATE ENDPOINT. THE STUDY ALSO USED MORE THAN THE MINIMUM NUMBER OF TREATMENT GROUPS AND INCLUDED A LONG POST-TREATMENT RECOVERY PERIOD. ALTHOUGH THE EXTENSIVE DATA BASE INCLUDES STUDIES COVERING ALL IMPORTANT TOXICOLOGIC ENDPOINTS AND SUPPORTS THE CHOICE OF CRITICAL EFFECT, IT IS ASSIGNED A MEDIUM CONFIDENCE LEVEL SINCE THER ARE NO ACCEPTED CHRONIC STUDIES. CONFIDENCE IN THE RFD IS RATED AT MEDIUM ONLY BECAUSE OF THE LACK OF A SENSITIVE MEASURE OF THE CRITICAL EFFECT FOR CHRONIC EXPOSURE.

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Below are some common definitions used in noncancer risk assessment. These definitions are for illustration only. Other terms are used in different organizations and countries.

Acceptable Daily Intake – An estimate of the dose resulting from daily exposure to a toxicant that is likely to be without deleterious effect even if continued exposure occurs over a lifetime.

Acute Exposure – One dose or multiple dose exposure occurring within a short time (<24 hours).

Adaptive Effect – Effects that enhance an organism's performance as a whole and/or its ability to withstand a challenge (i.e., homeostatic mechanism). An increase in hepatic smooth endoplasmic reticulum is an example of an adaptive effect, if hepatic metabolism reduces the chemical's toxicity.

Adverse Effect – A biochemical change, functional impairment, or pathological lesion which impairs performance and reduces the ability of the organism to respond to additional challenge.

Chronic Exposure – Multiple or continuous exposures occurring over an extended period of time, or a significant fraction of the animal's or the individual's lifetime.

Compensatory Effect – Effects that maintain overall function without enhancement or significant cost. Increased respiration due to metabolic acidosis is an example of a compensatory effect.

Critical Effect – A chemical often elicits more than one toxic effect, even in one species, or in tests of the same or different duration. The critical effects are the first adverse effects or their known precursors that occur as dose rate increases. The critical effects may change among toxicity studies of different durations, may be influenced by toxicity in other organs, and may differ depending on the availability of data on the shape of the dose-response curve.

Frank-Effect Level (FEL) – The exposure level which produces unmistakable adverse effects, such as reversible or irreversible functional impairment or mortality, at a statistically or biologically significant increase in frequency or severity between an exposed population and its appropriate control.

Health Hazard (types of) (Adapted in part from Klaasen et al., 1986) –

1. **Acute Toxicity** – Another term used to describe immediate toxicity (see below). Its use is associated with toxic effects that are severe (e.g., mortality) in contrast to the term "subacute toxicity", which is associated with toxic effects that are less severe. The term "acute toxicity" is often confused with that of acute exposure.

2. **Allergic Reaction** – Adverse reaction to a chemical resulting from previous sensitization to that chemical or to a structurally similar one.

3. **Chronic Toxicity** – The term "chronic toxicity" refers to effects that persist over a long period of time whether or not they occur immediately or are delayed. The term "chronic toxicity" is often confused with that of chronic exposure and is often used to describe delayed toxicity (see below).

4. **Idiosyncratic Reaction** – A genetically determined abnormal reactivity to a chemical.

5. **Immediate versus Delayed Toxicity** – Immediate effects occur or develop rapidly after a single administration of a substance, while delayed effects are those that occur after the lapse of some time. These effects have also been referred to as acute and chronic, respectively.

6. **Reversible versus Irreversible Toxicity** – Reversible toxic effects are those that can be repaired, usually by a specific tissue's ability to regenerate or mend itself after chemical exposure, while irreversible toxic effects are those that cannot be repaired.

7. **Local versus Systemic Toxicity** – Local effects refer to those that occur at the site of entry (e.g., lungs, stomach) of a toxicant into the body; systemic effects are those that are elicited after absorption and distribution of the toxicant from its entry point to a distant site.

Lowest-Observed-Adverse-Effect Level (LOAEL) – The lowest exposure level at which there are statistically or biologically significant increases in frequency or severity of adverse effects between the exposed population and its appropriate control group.

Modifying Factor (MF) – An uncertainty factor that is greater than zero and less than or equal to 10; the magnitude of the MF depends upon the professional assessment of scientific uncertainties of the study and data base not explicitly treated with the standard uncertainty factors (e.g., the number of animals tested); the default value for the MF is 1.

No-Observed-Adverse-Effect Level (NOAEL) – An exposure level at which there are no statistically or biologically significant increases in the frequency or severity of adverse effects between the exposed population and its appropriate control; *some effects may be produced at this level, but they are not considered as adverse, nor precursors to specific adverse effects*. In an experiment with several NOAELs, the regulatory focus is primarily on the NOAEL seen at the highest dose. This leads to the common usage of the term NOAEL to mean the highest exposure without adverse effect.

No-Observed-Effect Level (NOEL) – An exposure level at which there are no statistically or biologically significant increases in the frequency or severity of *any* effect between the exposed population and its appropriate control.

Reference Concentration (RfC) – An estimate (with uncertainty spanning perhaps an order of magnitude) of a continuous inhalation exposure to the human population (including sensitive subgroups) that is likely to be without appreciable risk of deleterious effects during a lifetime.

Reference Dose (RfD) – An estimate (with uncertainty spanning perhaps an order of magnitude) of a daily exposure to the human population (including sensitive subgroups) that is likely to be without appreciable risk of deleterious effects during a lifetime.

Subchronic Exposure – Multiple or continuous exposures occurring usually over 3 months.

Uncertainty Factor (UF) – One of several, generally 10-fold, factors used in operationally deriving the Reference Dose (RfD) from experimental data. UFs are intended to account for 1) the variation in sensitivity among the members of the human population; 2) the uncertainty in extrapolating animal data to the case of humans; 3) the uncertainty in extrapolating from data obtained in a study that is of less-than-lifetime exposure; 4) the uncertainty in using LOAEL data rather than NOAEL data; and 5) the inability of any single study to address adequately all possible adverse outcomes in man.

How Toxicity Data are Used in the Process of Hazard Identification and Dose-Response Assessment

Part 2 : Cancer Risk Assessment

Cancer Risk Assessment - Critical Issues

- Confidence chemical is an animal/human carcinogen
- Extrapolation across species (animal to human)
- Extrapolation within species (human to human)
- Extrapolation from high to low doses

Confidence Chemical Is an Animal/Human Carcinogen

- EPA 1986 Scheme - Weight of Evidence

  A (definite) >B (probable) >C (possible) >D (not known) >E (not)

  Based on number of species, tumor sites, etc.

  Limited consideration of mechanisms

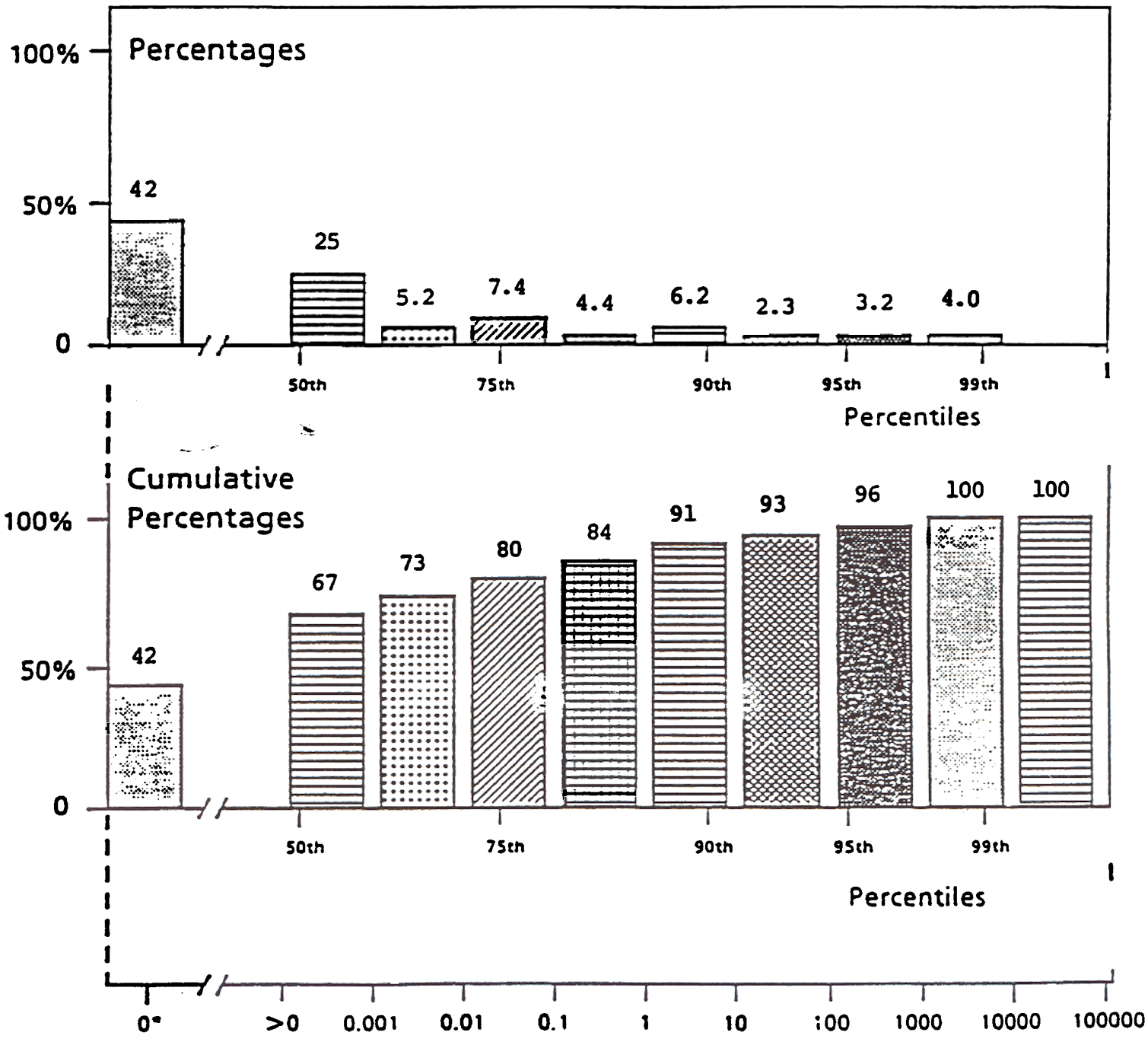
  Unintended consequences
- IARC/NTP - strength of evidence

Alternative Classification Approaches

- Ashby *et al.* (1990)
  - 8 categories - in-depth consideration of mechanisms
  - Allows for carcinogenicity in animals, not in humans
- Harvard Center for Risk Analysis (1992)

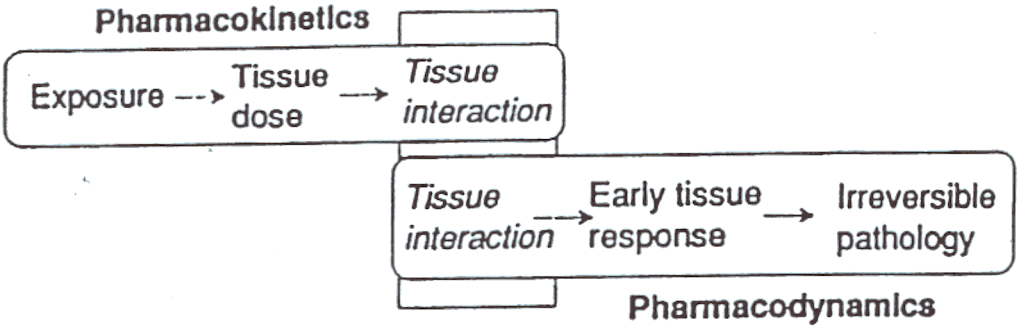
  Focuses on dose-response
  - Probabilistic description of dose-response relationships (including no response)

Predicted Cancer Incidence at 2.28 ppb Formaldehyde



(HCRA, 1992)

Extrapolation Issues



(Conolly and Andersen, 1991)

Extrapolation Across Species

Body weight *vs.* body weight $^{2/3}$ *vs.* body weight $^{3/4}$ (EPA/CPSC/FDA workgroup)

- Physiologically based pharmacokinetic (PBPK) models to estimate integrated dose of toxic metabolite
- Species-specific responses - *e.g.*, male rat kidney tumors (d-limonene, unleaded gasoline)

17
40

Use of PBPK Model - Plausible Upper Limit Estimate on Potency $(mg/kg/d)^{-1}$

Chemical	Standard Method	PBPK Model
Ethylene oxide	0.028	0.019
Butadiene	0.098	0.032
Perchloroethylene	0.0033	0.013

(Hattis, 1991)

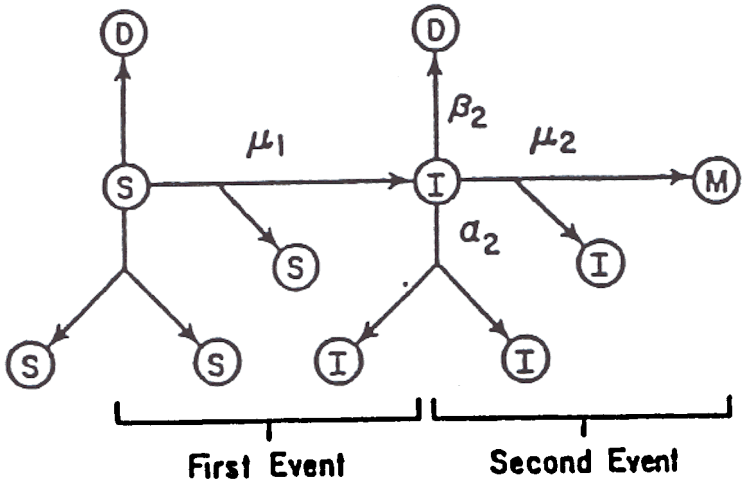
Extrapolation Within Species

- Inter-human variability in susceptibility - prior exposure, gender, genetics
- Poorly addressed in cancer risk assessment

Extrapolation from High to Low Dose

- PBPK model - saturation of activating/deactivating pathway
- **Dose-dependent mechanisms - *e.g.*, proliferation and genotoxicity at high doses *vs.* genotoxicity at low doses**
- Use of biologically based models *vs.* default models (*i.e.*, LMS)

Biologically Based Model for Carcinogenesis



S = normal stem cell
D = differentiated or dead cell
I = intermediate cell
M = malignant cell

μ_1, μ_2, d_2, β_2 = rate constants

(Moolgavkar, 1986)

Use of Biologically Based Model for Chloroform Risk Assessment

Daily Human Dose Associated with 10^{-6} Cancer Risk
Virtual Safe Dose (VSD)

Assumed Mechanism	Exponential Model VSD (mg/d)	Geometric Model VSD (mg/d)	Linearized Multistage Model VSD (mg/d)
Promotion only	950	730	-
Mutation only	0.011	0.011	0.30

(Bogen, 1990)

Additional Reading:

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