PDA Training Course Extractables & Leachables

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Toxicological Safety Evaluations of Extractables & Leachables

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PDA Parenteral Drug Association

Nelson Labs A Sotera Health company

CONNECTING PEOPLE SCIENCE AND REGULATION®

Topics Covered

- Basic Toxicological Principles
- Key Toxicological Endpoints
- Extractables and Leachables Qualification
- Best Practice Conclusions

Basic Toxicological Principles

Basic Toxicological Principles

"The Dose Makes the Poison"

Paracelsus, Swiss MD (1492-1541)

Hypothesis:

"All compounds are toxic, but below a certain dose – they are NOT"
"Only the dose makes the poison"

➔ Concept of **NOAEL**

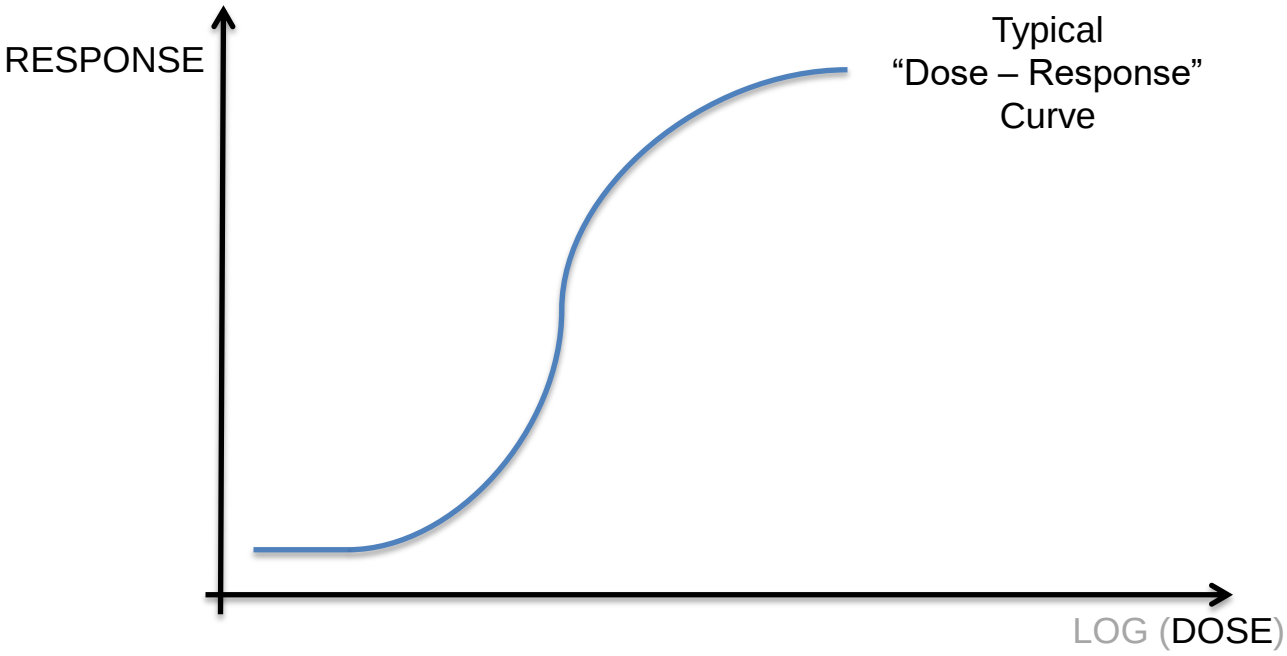
No **O**bserved **A**dverse **E**ffect **L**evel

Basic Toxicological Principles

THE DOSE-RESPONSE RELATIONSHIP

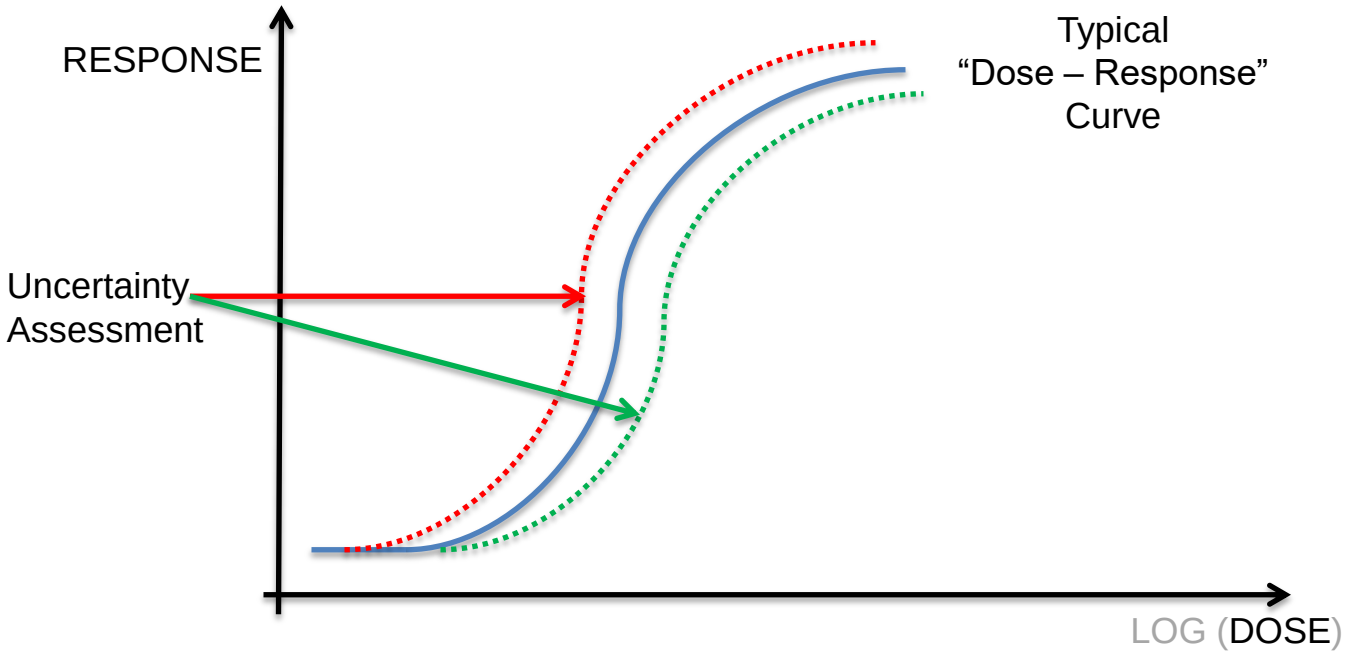
Basic Toxicological Principles

THE DOSE-RESPONSE RELATIONSHIP



Basic Toxicological Principles

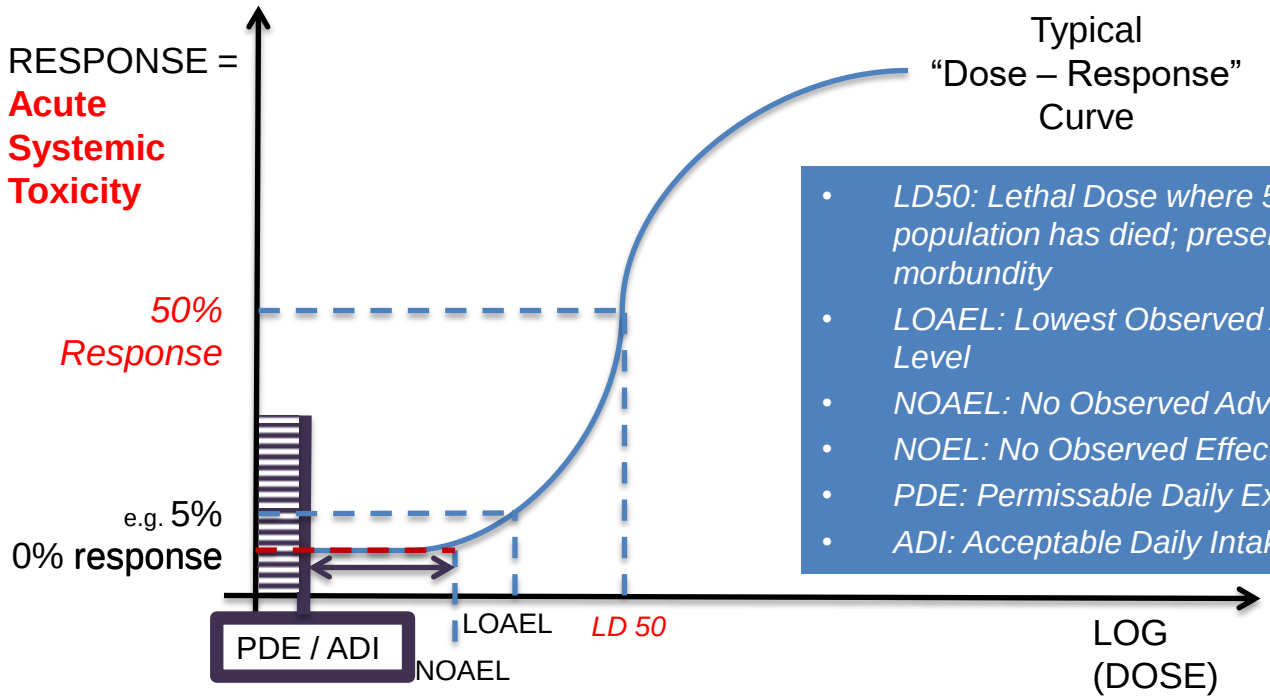
THE DOSE-RESPONSE RELATIONSHIP



Basic Toxicological Principles

THE DOSE-RESPONSE RELATIONSHIP

EXAMPLE: **ACCUTE SYSTEMIC TOXICITY**



- *LD50: Lethal Dose where 50% of the population has died; presents mortality & morbundity*
- *LOAEL: Lowest Observed Adverse Effect Level*
- *NOAEL: No Observed Adverse Effect Level*
- *NOEL: No Observed Effect Level*
- *PDE: Permissable Daily Exposure*
- *ADI: Acceptable Daily Intake*

Key Toxicological Endpoints

Toxicological endpoints to be considered (non – limitative):

A. Systemic Toxicity → "Dose – Response" Curve (Eg. LD50, NOAEL, LOAEL, ...)

B. Genotoxicity

C. Irritation

D. Sensitization

E. Reproduction Toxicity

F. Carcinogenicity

The "BIG FIVE"

A. Systemic Toxicity

= estimation of the human hazard potential of a substance by determining its systemic toxicity in a test system (animals)

- **Acute systemic toxicity**:
  - Single dose exposure (<24 hrs)
  - Major toxicity 1 or 2 organs
  - LD50 value
- **(sub)Chronic Systemic toxicity:**
  - Repeated exposure
  - negative control; LOW-; MID- and HIGH- dose group
  - Low dose ~ NOEL or NOAEL or LOAEL

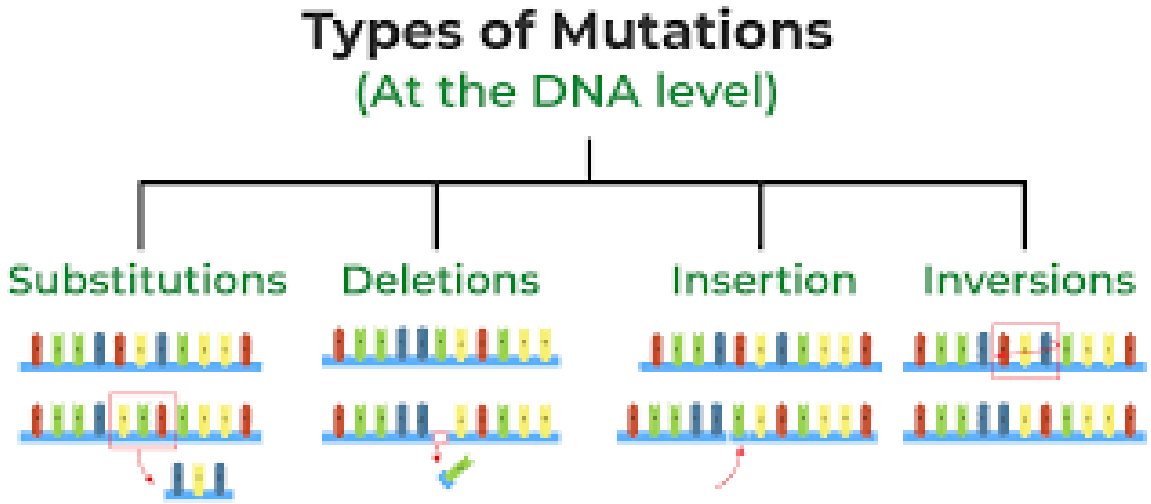
Source: alttox.org

Ex. OECD Test No. 407: Repeated Dose 28-day Oral Toxicity Study in Rodents

B. Genotoxicity

= genetic damage.

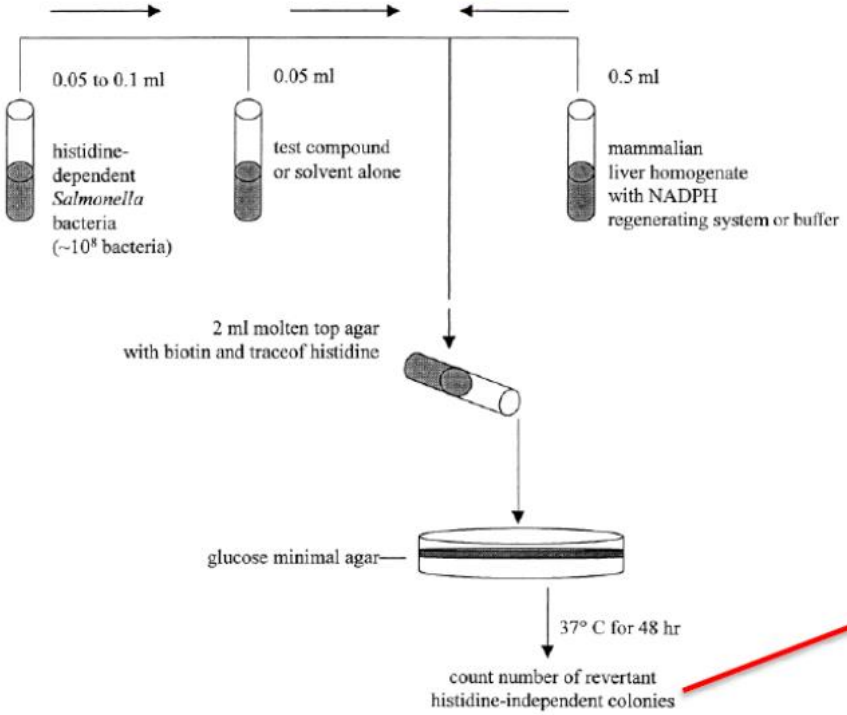
- DNA Level = mutagenicity
- Chromosomal Level = Clastogenicity and Aneugenicity

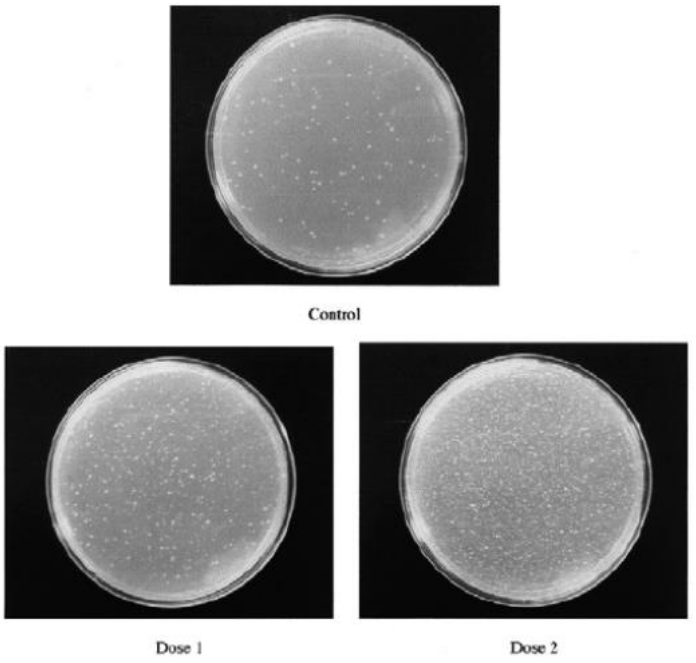


Ex.OECD 471: Bacterial Reverse Mutation Test (AMES)

B. Genotoxicity

- **Ames Assay**





Mortelmans K., Zeiger E. (2000) *Mut. Res.* 455:29-60

C. Irritation/ Corrosion

= localized toxic effects resulting from a topical exposure of the skin to a substance

- **Skin irritation**
  = Reversible damage
  Eg. Rash development

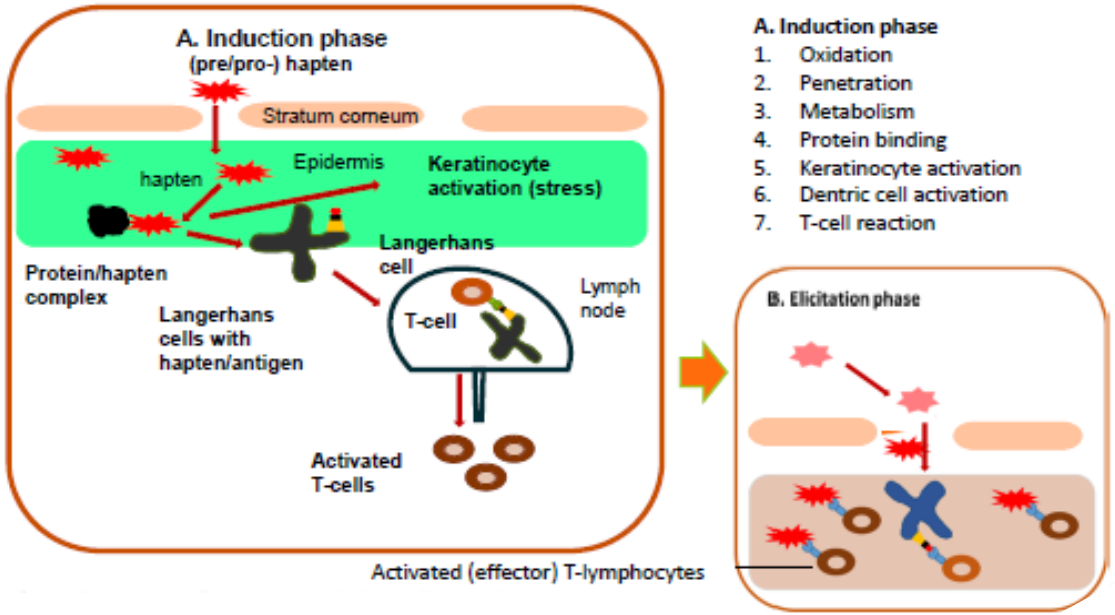
- **Skin corrosion**
  = Irreversible damage
  Eg. necrosis through the epidermis and into the dermis

OECD 404 Skin Irritation Test

Source: alttox.org

D. Sensitization

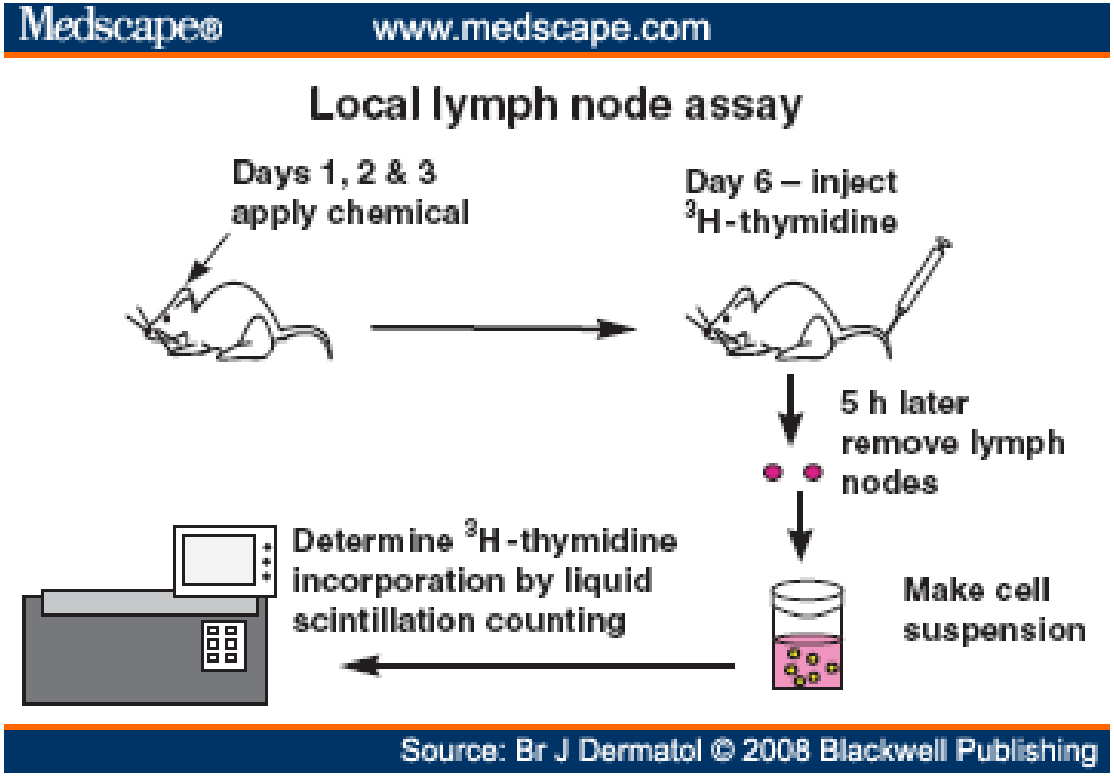
= induction of an **allergic response following (repeat) skin contact**".



- **Allergic Responses: Often Dose Independent!!**

OECD 429 Local Lymphnode Assay (LLNA)

D. Sensitization



Stimulation Index (SI) versus control (≥3 = positive)
EC3 value = []% at which SI = 3 (➔ weak, moderate, strong, extreme sensitizers)

E. Reproductive Toxicity

Reproductive toxicity includes the toxic effects of a substance on the reproductive ability of an organism and the development of its offspring (teratogenicity).

Reproductive toxicity
= adverse effects (of chemicals) on sexual function and fertility in adult males and females, as well as developmental toxicity to the offspring during pregnancy

Developmental toxicity
= adverse effects induced during pregnancy, or as a result of parental exposure (i.e. via breast feeding)…manifested at any point in the life span of the organism

Source: alttox.org

F. Carcinogenicity

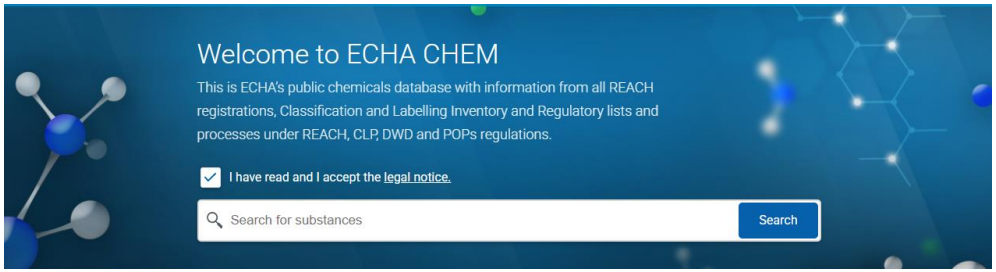
Carcinogen
= a chemical substance or a mixture of chemical substances which **induce cancer** or **increase its incidence**".

An alternate definition is that ***carcinogenic substances*** are ones that "**induce tumors** (benign or malignant), **increase** their **incidence or malignancy**, or **shorten** the **time** to **tumor occurrence** when they are inhaled, injected, dermally applied, or ingested

- *Genotoxic:* directly altering the genetic material
- *non-genotoxic: secondary mechanism not related to direct gene damage*.

1-2Y Carcinogenicity study: determine Toxic Dose 50% or TD50 at which exposure 50% of the test animals develop tumors

Key Toxicological Endpoints



http://echa.europa.eu/
https://chem.echa.europa.eu/
http://www.epa.gov/hpvis/
http://webnet.oecd.org/hpv/
http://www.inchem.org/
http://ntpapps.niehs.nih.gov/ntp_tox/index.cfm

Extractables and Leachables Qualification

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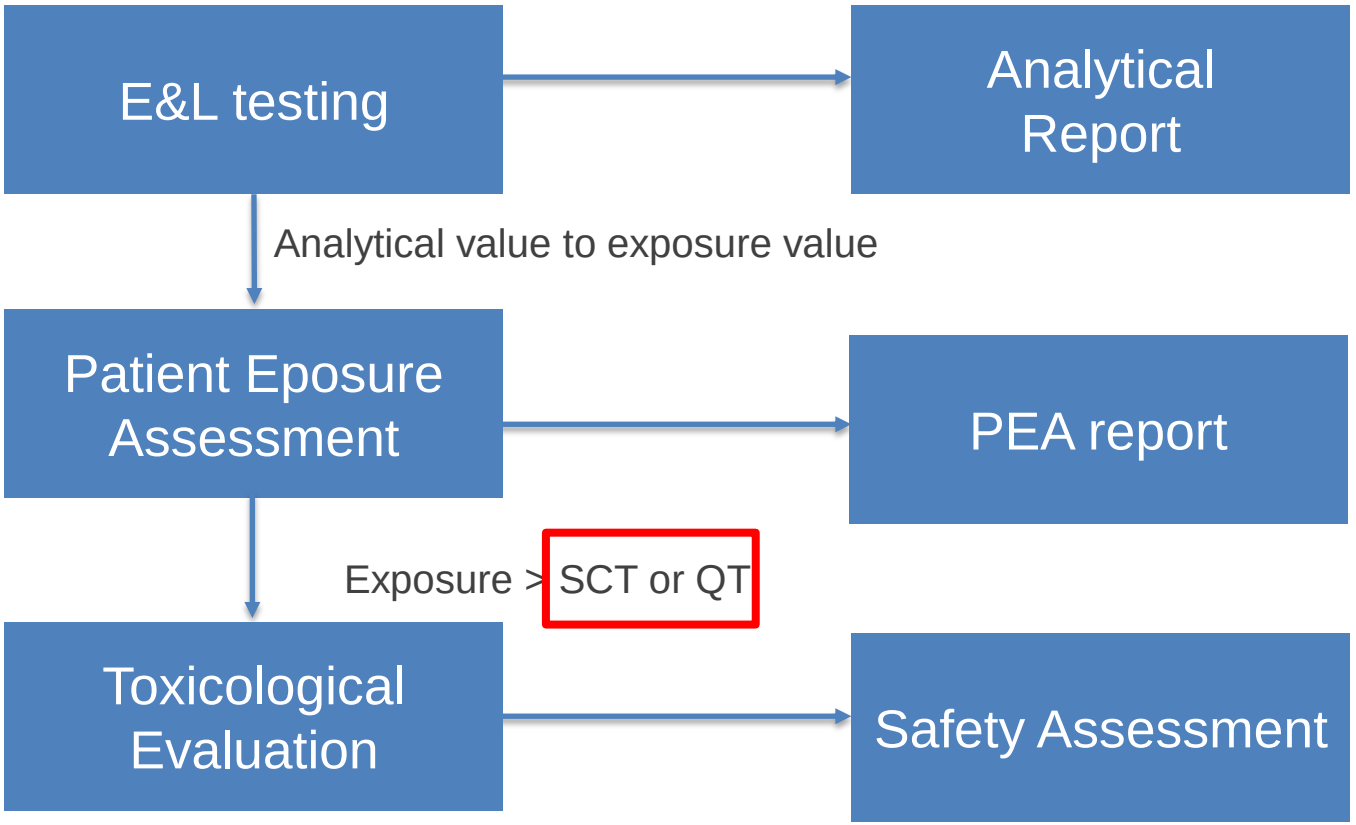


➔ Hazard assessment: PDE determination

➔ Risk assessment

Safety Limits

A. Mutagenic impurities – ICH M7
B. Sensitizers and irritants – PQRI
C. Extractables and Leachables – ICH Q3E

A. Mutagenic impurities – ICH M7

ICH M7: Assessment & Control of DNA Reactive (Mutagenic) Impurities in Pharmaceuticals to Limit Potential Carcinogenic Risk

➔Mutagenic impurities:

- Production of transmissible genetic alterations from cell to cell or generation to generation
- Can lead to cancer

Purpose: Provide a framework for

- Identification
- Categorization
- Quantification
- Control

of mutagenic impurities to limit potential carcinogenic risk

A. Mutagenic impurities

Threshold of Toxicological Concern (TTC)

- Daily exposure to **1.5 µg/day** for most (genotoxic) carcinogens **not likely to exceed lifetime cancer risk** of 1 in 10^5

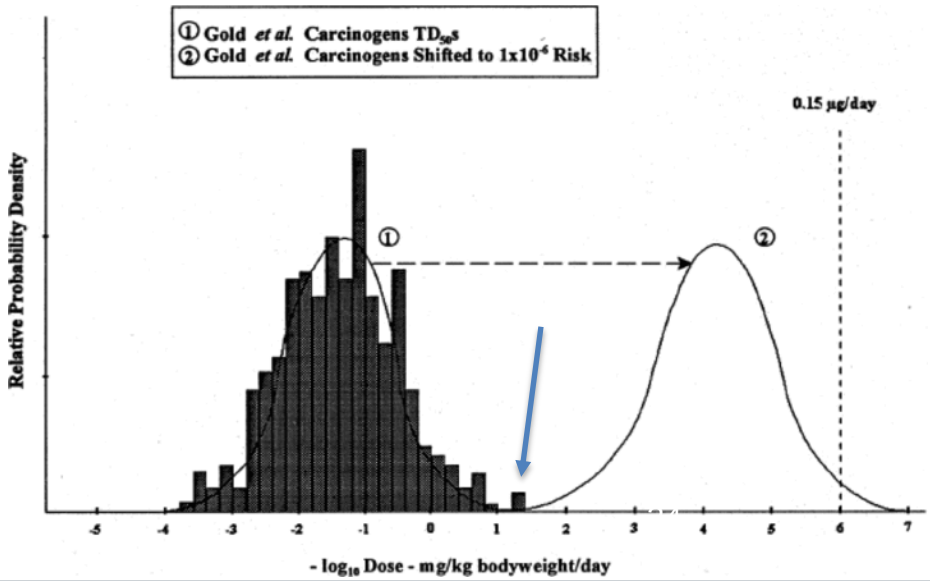
  = Acceptable risk

Exception: Cohorts of Concern
(include aflatoxin-like, azoxy and N-nitroso compounds
– need case-by-case assessment)

- Staged TTC Approach

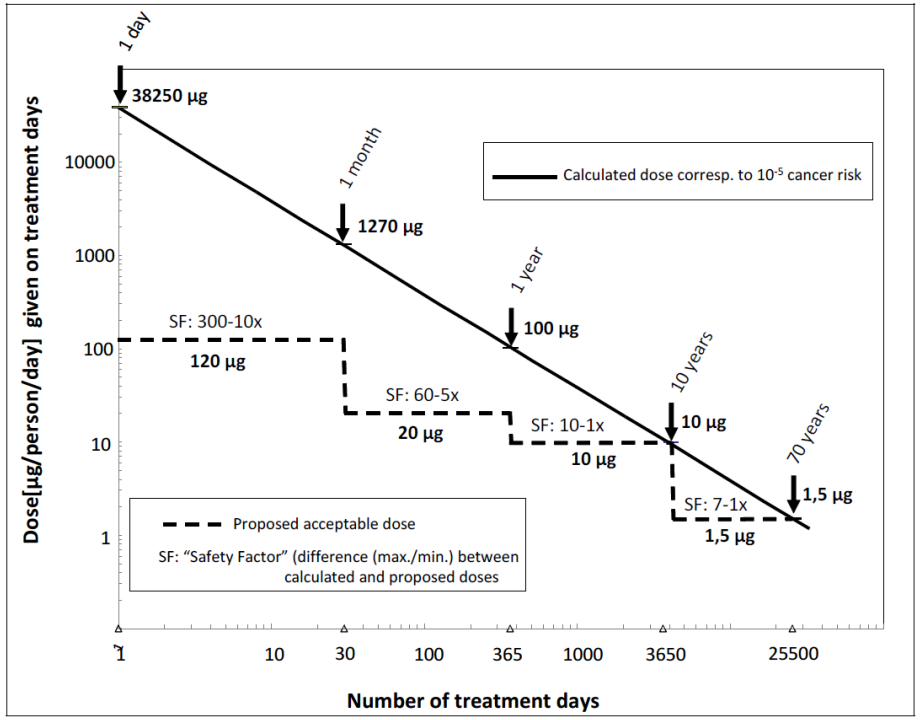
– based on Haber's Rule

$C \times t = k$ With C = Concentration
t = time
k = constant



A. Mutagenic impurities

Duration of treatment	≤1 month	>1 -12 months	>1 -10 years	>10 years
Daily intake (µg/day)	120	20	10	1.5



Uniformly distributed over total Number of exposure days

HABER's RULE:

$C_1t_1=C_2t_2$

Eg. 1,5 µg/day x 25.550 days
= 38,3 mg (x 1 day)

B. Non-Mutagenic impurities

PQRI: **SAFETY CONCERN THRESHOLD (SCT)**

"Threshold below which a leachable would have a dose so low as to present negligible safety concerns from carcinogenic and non-carcinogenic toxic effects"

PQRI OINDP

Tox Endpoint	Others	Sensitizer & Irritant	Carcinogen
Class	Class I	Class II	Class III
Threshold Level (µg/day)	PDE	5	0.15

PQRI PDP

Tox Endpoint	Others	Sensitizer & Irritant	Carcinogen
Class	Class I	Class II	Class III
Threshold Level (µg/day)	PDE	5	1.5

Threshold Recommendations

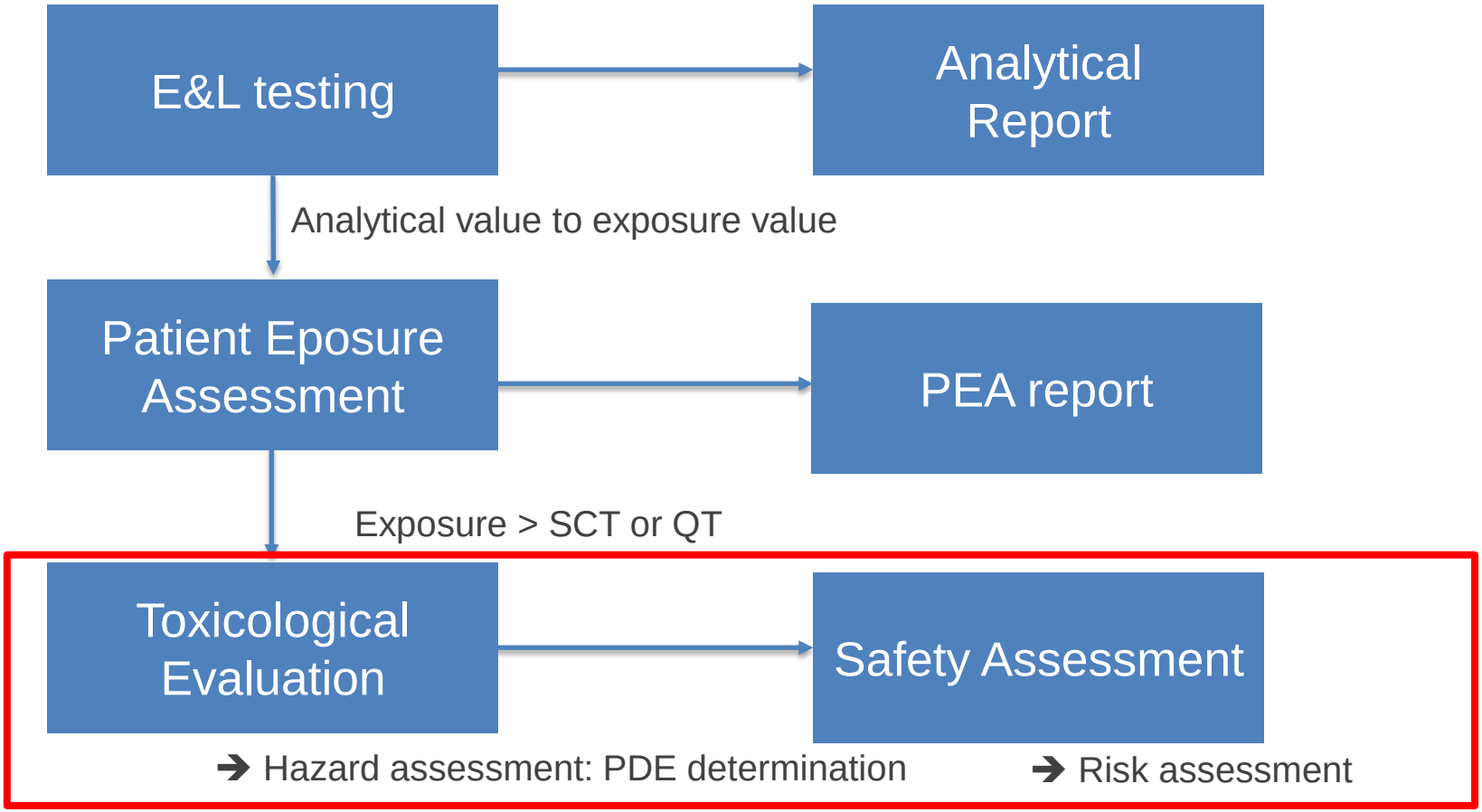
Acceptable Daily Intake (µg/day)				
Toxicological Endpoint	Duration of Therapy			
	≤ 1 month	1-12 months	1-10 years	> 10 years
Mutagenitic (TTC, SCT)	120	20	10	1.5
Sensitization – Irritation	5	5	5	5
General	PDE	PDE	PDE	PDE
ICH Q3E (draft)	26	12	12	12

Staged TTC approach ICH M7

Conclusion:

- The **need to have the correct chemical structure** & Identity above the Q.T.
  - *For **Chronic** Treatments: Q.T. = **1,5** µg/day*
  - *For **All other** treatments: Q.T. = **5** µg/day*
- **Compound Identity** can make the **link to** the **toxicology** (carcinogen or sensitizer?)
- As such, the **Qualification Threshold** (QT) **becomes** an **Identification Threshold**!
- As it is a requirement for **Leachables**, a **screening step** should be built into the Leachables Study Design.

Extractables and Leachables Qualification



General Framework

Exposure Assessment

- Extractable/leachable study well designed?
- Frequency of dosing: less-then-lifetime vs. Chronic exposure
- Route of Exposure (IV, SC, IM, dermal, ...)
- Patient exposure (µg/day) (= concentration ext/lea * MDD)

Risk Assessment

- Which thresholds do we use?
  - Generic TTC (lifetime, staged, less-than-lifetime) or QT
  - Compound Specific PDE
- Safety Margin

= safety threshold (µg/day) / Patient exposure (µg/day) > 1

Hazard Assessment

- Literature research
  - Classifications
  - Experimental Data
- Prediction Models

→ Mostly no or limited data available

Exclude mutagenicity & sensitisation potential

In parallel or Stepwise

Prediction Models

QSAR systems
= **Q**ualitative **S**tructure **A**ctivity **R**elationship Assessment

Rule based

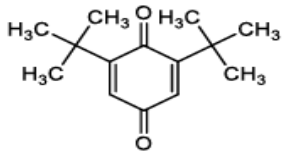
DEREK = Deductive Estimation of Risk from Existing Knowlegde
E.g. Mutagenicity is PLAUSIBLE / PROBABLE …

Statistically based

Multicase, LeadScope, Sarah
E.g. < 40 (negative); 40-60 (inconclusive); >60 (positive)

Prediction Models

2,6-Di-*tert*-butyl-2,5-cyclohexadiene-1,4-dione
2,6-Bis-(1,1-dimethylethyl)-2,5-cyclohexadiene-1,4-dione
2,6-Di-*tert*-butyl-1,4-benzoquinone



[719-22-2] $C_{14}H_{20}O_2$ 220.31

FIT Screening Evaluation:
Cramer Classification: Class II
Derek predictions (Reasoning summary and alerts found):

- Chromosome damage in vitro in bacterium is IMPOSSIBLE; 1,4-Benzoquinone or -naphthoquinone
- Chromosome damage in vitro in human is PLAUSIBLE; 1,4-Benzoquinone or -naphthoquinone
- Chromosome damage in vitro in mammal is PLAUSIBLE; 1,4-Benzoquinone or -naphthoquinone
- Chromosome damage in vivo in bacterium is IMPOSSIBLE; 1,4-Benzoquinone or -naphthoquinone
- Chromosome damage in vivo in human is EQUIVOCAL; 1,4-Benzoquinone or -naphthoquinone
- Chromosome damage in vivo in mammal is EQUIVOCAL; 1,4-Benzoquinone or -naphthoquinone
- Mitochondrial dysfunction in bacterium is IMPOSSIBLE; 1,4-Benzoquinone
- Mitochondrial dysfunction in human is EQUIVOCAL; 1,4-Benzoquinone
- Mitochondrial dysfunction in mammal is EQUIVOCAL; 1,4-Benzoquinone
- Mutagenicity in vitro in bacterium is PLAUSIBLE; Quinone, precursor or analogue
- Skin sensitisation HPC in bacterium is IMPOSSIBLE; Class 2: Michael acceptor
- Skin sensitisation HPC in human is PLAUSIBLE; Class 2: Michael acceptor
- Skin sensitisation HPC in mammal is PLAUSIBLE; Class 2: Michael acceptor
- Skin sensitisation in bacterium is IMPOSSIBLE; Quinone, quinoneimine or precursor
- Skin sensitisation in human is PLAUSIBLE; Quinone, quinoneimine or precursor
- Skin sensitisation in mammal is PLAUSIBLE; Quinone, quinoneimine or precursor

FIT Screening Classification: Class III
Suggested Threshold Level: 1.5 µg/day

Compound Specific PDE

= Permissible Daily Exposure

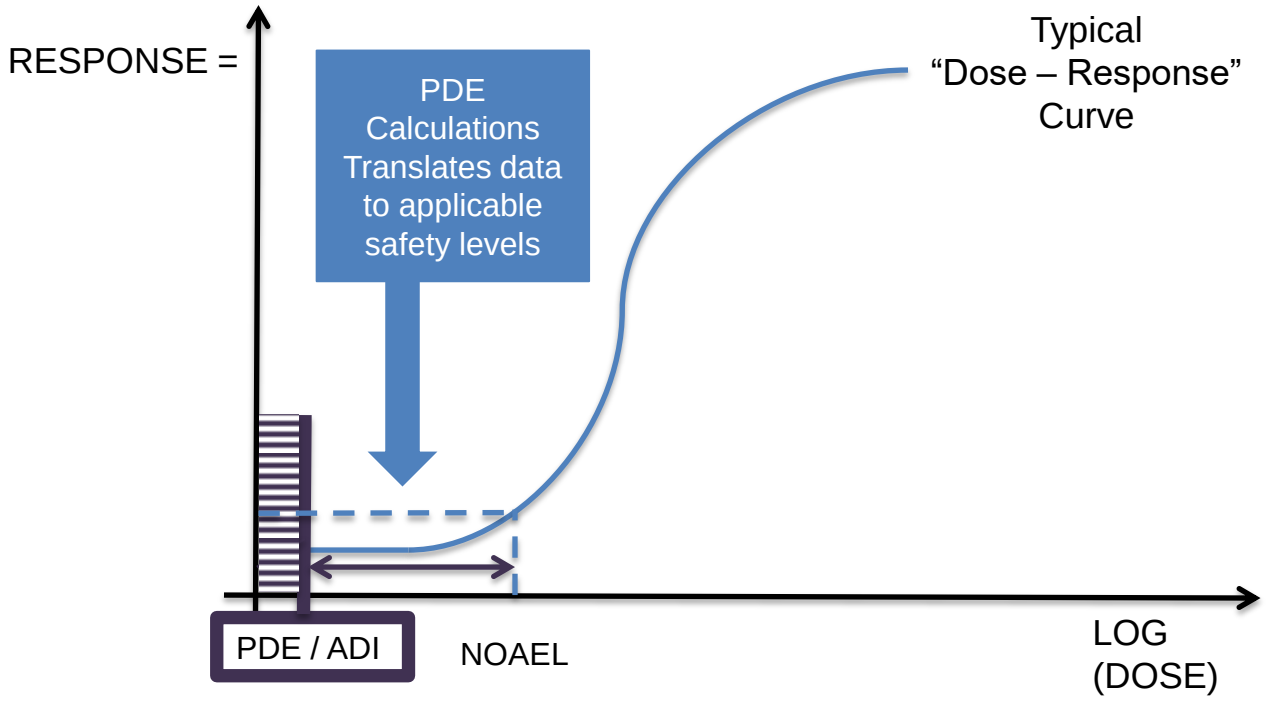
$$PDE = \frac{\text{NO(A)EL x Weight Adjustment}}{\text{F1 x F2 x F3 x F4 x F5}}$$

F1 = Variation between Species
F2 = for Variation between individual Humans
F3 = Short Duration in Animals to Chronical Human Exposure
F4 = Teratogenicity, Neurotoxicity and non-genotoxic carcinogens
F5 = applied if the no-effect level was not established (10 for using LOAEL)
F6 = route of administration: factor 10 from oral to I.V
F7 = read across (ICH Q3E draft).

REMARK: NEVER USE LD50 TO CALCULATE A PDE!

Basic Toxicological Principles

THE DOSE-RESPONSE RELATIONSHIP



Example

Staged TTC approach can be applied

Duration of treatment	≤1 month	>1 -12 months	>1 -10 years	>10 years
Daily intake (µg/day)	120	20	10	1.5

ex. Patient exposure calculation

- MDD = 6 units/day
- Accumulative exposure = 140 days (less-than-life-time)

Organic compound	Highest observed value in E&L study (µg/unit)	Patient exposure (µg/day)*	Safety Threshold (µg/day)	Margin of safety (MOS)**	PDE (µg/day)	Margin of safety (MOS)**
Compound 1	0.81	4.86	5	1.03	-	-
Compound 2	0.16	0.96	5	5.21	-	-
Compound 3	0.32	1.92	5	2.60	-	-
Compound 4	**3.4**	**20.4**	**5**	**0.25**	**1378**	**67.5**
Compound 5	0.26	1.56	5	3.21	-	-
Compound 6	**2.1**	**12.6**	**5**	**0.40**	**35448**	**2813**
Compound 7	**8.9**	**53.4**	**5**	**0.09**	**1418**	**26.5**

*Patient exposure (µg/day) = Highest observed extractable level (µg/unit) x maximum daily dose (i.e. 6 units /day);

**MOS = Safety threshold (µg/day) / patient exposure (µg/day)

Best Practice Conclusions

Best Practice Conclusions

- The Dose Makes the Poision – Paracelsus
- NOAEL/NOEL Levels in Systemic Toxicity testing allow to calculate PDE levels when not:
  - Mutagenic – carcinogenic
  - Sensitizing or irritating
- Conservative approach taken for Mutagenic Impurities
  - Use of Linear extrapolation to 1 in 100,000 risk, used to establish TTC – lifetime limit of 1.5 ug/day.
  - Staged Approach (based upon Haber's Rule) can be used where the identified compound is identified to be a potential carcinogen, mutagen or genotoxic compound *(and compound is not sensitizer/irritant)*
    - This concept CANNOT be used as an IDENTIFICATION THRESHOLD in Extractables & Leachables (concern for sensitizers)
    - If a compound has Actual Toxicity Data on Carcinogenicity/Mutagenicity, USE AVAILABLE DATA, instead of generic approach
      - Often, this will allow you to increase the level of concern for the compound.

Best Practice Conclusions

- Final Toxicological Assessment needs to be done on the “quantitative” Leachable results
- Leave toxicology to toxicologists; all assessments must be verified by a certified Toxicologist.