Toxicology – 2

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What is Toxicology?

- Toxicology is the scientific study of adverse effects that occurs in living organisms due to chemicals.
- it involves observing and reporting symptoms, mechanisms, detection and treatments of toxic substances, in particular relation to the poisoning of humans.

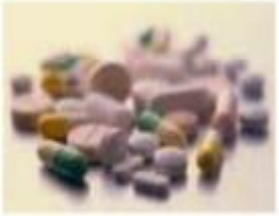
✓Why study toxicology???

- Benefit –risk ratio can be calculated

- Prediction of therapeutic index

$$\text{Therapeutic index} = \frac{\text{Maximum tolerated dose}}{\text{Minimum curative dose}}$$

- Smaller ratio, better safety of the drug



In Vitro Toxicology

- Screening
  - Cytotoxicity
  - Protein binding
  - CYP inhibition/induction
  - Membrane permeability
  - Metabolic stability
  - Interspecies comparison

In Vitro Toxicology
Dermal or Ocular Toxicity

- Replace in vivo tests such as Dermal Corrosion, Skin Irritation, Draize Eye Irritancy

- Many tests now available in kit form

- Example: EpiDerm
  - Normal human epidermal keratinocytes
  - Cultured on a permeable polycarbonate membrane
  - Stratified, highly differentiated, model of human epidermis
  - Metabolically and mitotically active cells organized into differentiated layers

In Vitro Toxicology
Immunotoxicity

- These are, by necessity, functional tests

- Examples
  - Cytokine release. Assess ability of a chemical to induce release of cytokines/chemokines. Can use cells from various sources, including peripheral blood cells. Many variations of this assay. TGN 1412
  - Drug-induced histamine release. Cremophor, polysorbate 80

In Vitro Toxicology
Metabolism and Kinetics

- Assess ability of a chemical to induce metabolism of specific substrates, including the chemical itself
- Information about metabolic pathways by which the chemical can be metabolized
- Information on production of toxic/reactive metabolites
- Interspecies comparisons
  - Can provide information on relevance of a particular animal model from a metabolic standpoint

1.4 Local toxicity

- Required when the new drug is proposed to be used by some special route (other than oral) in humans.
- Applied to an appropriate site (e.g., skin or vaginal mucous membrane) to determine local effects in a suitable species.
- Typical study designs for these studies should include three dose levels and untreated and/ or vehicle control, preferably use of 2 species

C. Local toxicity studies

- Required when drug is administered by special route (other than oral) in humans

- Study design:
  - 2 species along with control used
  - Dose dependent on dose escalating studies
  - 3 dose levels

Local toxicity

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- Dermal toxicity study
- Photo-allergy or dermal photo-toxicity
- Vaginal Toxicity Test
- Rectal Tolerance Test
- Parentral Drugs
- Ocular toxicity studies
- Inhalation

Types of local toxicity studies

- **Dermal toxicity studies**

  - Rats & Rabbit
  - Local signs (erythema, oedema), histological examination

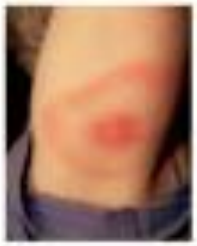
- **Dermal photo-toxicity studies**
  - Guinea pig
  - Used in treatment of leucoderma
  - Examination of erythema & oedema formation

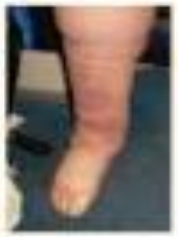
- **Vaginal toxicity studies**
  - Rabbit or Dog
  - Observation of swelling, histopathology of vaginal wall

- **Rectal tolerance studies**
  - Rabbit or Dog
  - Signs of pain, blood or mucous, histology examination of rectal mucosa

D. Allergenicity/hypersensitivity toxicology studies

- Guinea Pig Maximization test
  - Determination of Maximum non irritant or minimum irritant dose
  - Evaluation of Erythema and oedema





- Local lymph node assay
  - Mice of one sex(either male or female)
  - Drug treatment given on ear skin
  - Auricular lymph node dissection after 5 days
  - Increase in 3h-thymidine used for evaluation

Genotoxicity

- Genotoxic compounds, shall be presumed to be trans-species carcinogens, implying a hazard to humans.
- Such compounds need not be Subjected to long-term carcinogenicity studies.
- However, if such a drug is intended to be administered for chronic illnesses or otherwise over a long period of time - a chronic toxicity study (up to one year) may be necessary to detect early tumorigenic effects.

E. Genotoxicity studies

- To detect early tumorigenic effects in cases of chronic illness

- In vitro tests:
  - Test for gene mutation in Bacteria
  - Cytogenetic evaluation of chromosomal damage in mammalian cells

    E.g.; *Ames's Salmonella Assay* detects increased number of aberrations in metaphase chromosomes
  - DNA strand breaks, DNA repair or recombination, Measurements of DNA adducts
- In vivo tests:
  - Chromosome damage in rodent hematopoietic cells

    E.g.; *Micronucleus Assay*

❖ CARCINOGENICITY STUDY

451 **PRINCIPLE** 451

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- **The test substance is administered daily in graduated doses**
- **Observed closely for signs of toxicity and for the development of neoplastic lesions.**
- **Died or are killed animals are necropsied and at the conclusion surviving animals are also killed and necropsied.**

ROUTES OF ADMINISTRATION

F. CARCINOGENICITY/ ONCOGENICITY STUDIES

- Life-time Bioassays
- *Carcinogenicity studies are performed on:*
- Drug used for >6 months or frequent intermittent use for chronic diseases
- Chemical structure of drug indicates carcinogenic potential
- Therapeutic class of drugs which have produced positive carcinogenicity

CONDUCT OF STUDY

- Group sizes of 50 animals/sex at each of 3 dose levels
- Control group is of double size
- Record for onset of tumor development
- Usually carried out for 24 months in rats and 18 months in mice (life span studies)

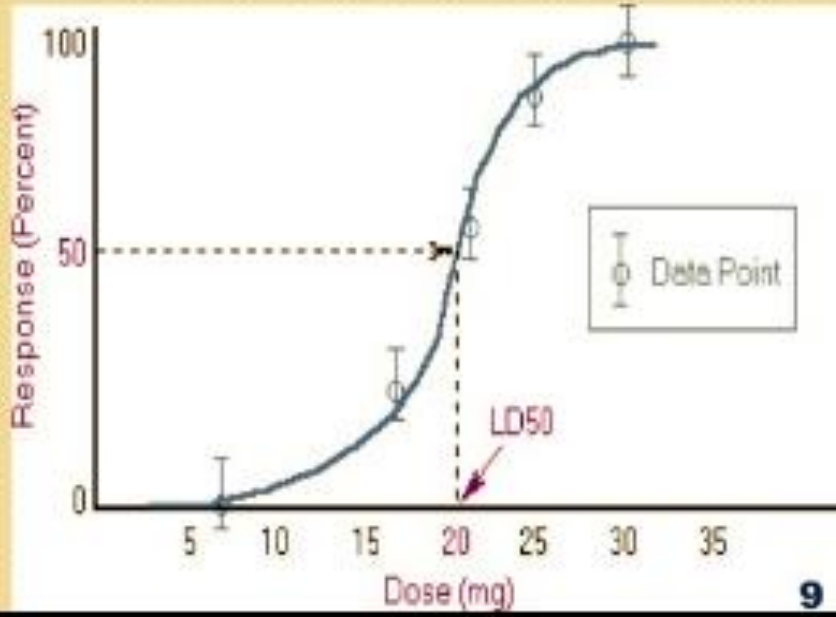
❖ WHAT IS LD_{50}? (P.A. Botham *et al.* 2004)

- **LD50 represents the individual dose required to kill 50 percent of a population of test animals. It is an index determination of medicine and poison's virulence. lower the LD50 dose, the more toxic the pesticide.**

❖ WHAT IS LC_{50}?

- **The concentrations of the chemical in air that kills 50% of the test animals during the observation period is the LC_{50} value. Other durations of exposure (versus the traditional 4 hours) may apply depending on specific laws.**

❖ LD50/LC50



What this means

higher LD_{50}/LC_{50} = less toxic

lower LD_{50}/LC_{50} = more toxic

THANK YOU!!