Preclinical studies needed in the development of human pharmaceutical drugs –
role of toxicology and risk assessment

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Preclinical studies needed in the development of human pharmaceutical drugs

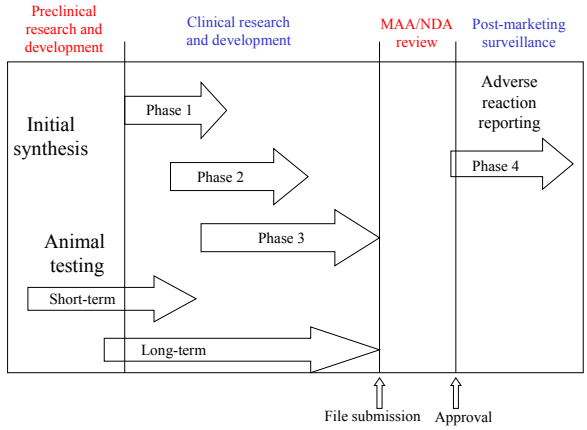
Pharmacology
- primary pharmacodynamic studies: studies on mode of action and/or effects of a substance to its desired therapeutic effects
- secondary pharmacodynamics: effects of a substance not related to its desired therapeutic target
- Safety pharmacology

Pharmacokinetics and metabolism

Toxicology

Factors affecting development strategy

- Guidelines
- Novelty
- Acceptable risk/Benefit
- Drug supply
- Expected market value
- Type of compound
- Ethics



Clinical studies

Phase 1
- Who: Normal volunteers or special populations (renal or hepatic impairment
- Why: Safety, biological effects, metabolism, kinetics

Phase 2
- Who: Selected patients
- Why: Therapeutic efficacy, dose range, kinetics, metabolism

Phase 3
- Who: Large sample of patients
- Why: Safety and efficacy. Compare to golden standard

Goals of non-clinical safety evaluation

- To be as sure as possible (within reasonable limits) that the products we develop are not harmful to man at clinically relevant doses
- To identify target organs or biomarkers that need to be followed-up in clinical studies
- To fulfill the regulatory requirements for registration of the products
- To improve our understanding of the biological effects of specific drugs
- To develop safer drugs in the future

Predictive value of non-clinical safety evaluation

From Greaves et al., Nature Drug Discovery, March 2003

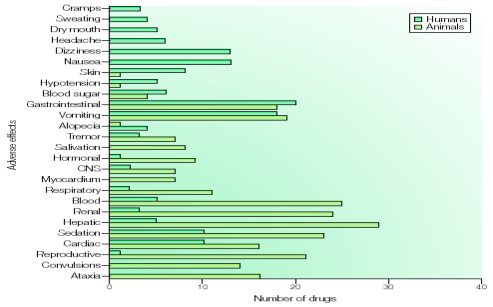


Figure 1 | **Animal and human toxicities of 45 drugs assessed by the Committee on Safety of Medicines in the United Kingdom during the eight or nine months prior to publication in 1978** (REF 11). Data are for drugs of diverse therapeutic classes, including several cardiovascular and central nervous system drugs but only one anticancer agent. The six uppermost adverse effects were observed in humans but not in animals; the two adverse effects at the bottom of the graph were observed in animals but not in humans. For most adverse effects there is a degree of over- or under-prediction. CNS, central nervous system.

Regulatory requirements in different regions in the world

- There were big differences between regulatory requirements between USA, Japan, and Europe.
- These differences are nowadays less.
  - Europe is more interested in mechanistic type of studies and more open for new types of studies/study designs
  - Japan: large focus on No-effect Levels, excipients and impurities.

International Commission on Harmonisation – guidelines (www.ICH.org)

S1A	Guideline on the need for carcinogenicity studies of pharmaceuticals
S1B	Testing for carcinogenicity in pharmaceuticals
S1C	Guidance for dose selection for carcinogenicity studies of pharmaceuticals (+ S1C(R): Addendum)
S2A	Genotoxicity: Specific aspects of regulatory genotoxicity tests
S2B	Genotoxicity: A standard battery for genotoxicity testing of pharmaceuticals
S3A	Toxicokinetics: Guidance on the assessment of systemic exposure in toxicity studies
S3B	Pharmacokinetics: Guidance for repeated dose tissue distribution studies

International Commission on Harmonisation – guidelines www.ICH.org

S4	Single dose toxicity tests
S4A	Duration of chronic toxicity testing in animals (rodent and non-rodent)
S5A	Detection of toxicity to reproduction for medicinal products
S5B(M)	Reproductive toxicology: Male fertility studies
S6	Safety studies for biotechnological products
S7A	Safety pharmacology studies for human pharmaceuticals
S7B	Safety pharmacology studies for assessing the potential for delayed ventricular repolarization (QT interval prolongation) by human pharmaceuticals
M3	Timing of pre-clinical studies in relation to clinical trials

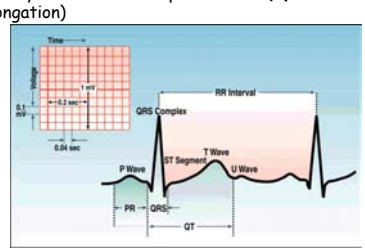
safety pharmacology

Safety pharmacology studies for human pharmaceuticals - ICH S7

- Core battery:
  - Effects on CNS system
    - motor activity, behavioural changes, coordination, sensory/motor reflexes. I.e. Modified Irwin test, Functional Observation test
  - Cardiovascular system
    - Blood pressure, heart rate and electrocardiogram
    - Preferably use conscious animals (telemetry models)
  - Respiratory system
    - Respiratory rate and tidal volume

Safety pharmacology studies for human pharmaceuticals - ICH S7

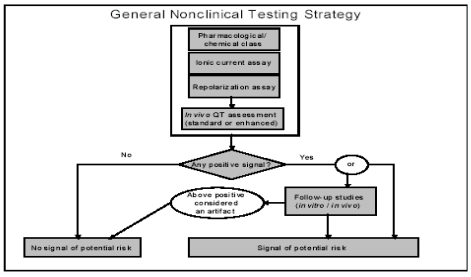
- ICH S7B: safety pharmacology for assessing the potential for delayed ventricular repolarization (QT interval prolongation)



Safety pharmacology studies for human pharmaceuticals - ICH S7

- QT interval is dependent on heart rate and should therefore be corrected fore heart rate (QTc)
- Long QT syndrome is QTc > 440 ms
- QT prolongation can lead to arrhytmias and/or Torsade de pointes
  - Antihistamines
  - Andidepressants
  - Anticonvulsants
  - Antiarrhytmic agents
  - etc

Safety pharmacology studies for human pharmaceuticals - ICH S7



Safety pharmacology studies for human pharmaceuticals - ICH S7

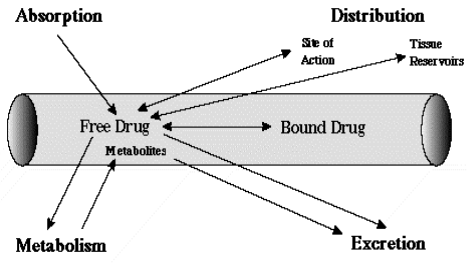
- HERG assay
  - HERG is a Human Ether-a-go-go gene Related Gene
  - transfected in human cells (human embryonal kidney cells)
  - Codes for a K+ channel
  - Measurements on individual cells by action potential clamp techniques

Pharmacokinetics and metabolism

Why is it important to know the pharmacokinetics of new drugs?

- Interpret toxic findings and side-effects
  - Maximum concentration and total exposure
- Predict drug-drug interactions
  - Are drugs metabolized by the same enzymes (P450 3A)?
- Predict influence of diseases on drug use
  - Liver diseases can lead to reduced drug metabolism
- Predict influence of age and gender on drug use
  - activity phase 1 drug metabolizing enzymes is reduced in elderly people
- Design dosing regimens

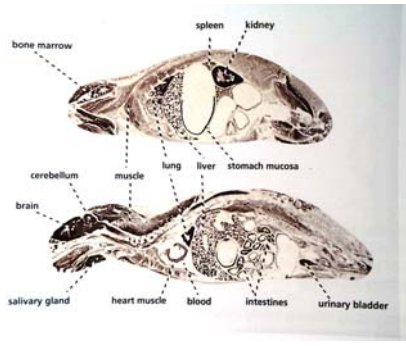
ADME



Quantitative whole-body autoradiography

- Test article labeled with an appropriate radioactive isotope
- Administration to animals
- Animals killed at different time points, and frozen
- Slices prepared by whole-body microtome
- Freeze-dried sections exposed to storage phosphor screens, which are scanned using a phosphor imager.

Quantitative whole-body autoradiography 14C glucose



Toxicology

Toxicology - How?

- Single dose toxicity
- Repeat dose toxicity
- Local tolerance
- Genetic toxicity, *in vitro* + *in vivo*
- Reproductive toxicity
- Carcinogenic potential
- Immunotoxicology
- Special studies

Single dose toxicity

ICH - 2 mammalian species

EU - 2 mammalian species

FDA - 2 mammalian species, justify if not dog

MWH - rodent + non-rodent other than rabbit

- DRF (to lethal / limit dose)
- Single dose via intended clinical route + i.v.
- Minimum 3 doses + control
- 14 Day observation
- Body weight
- Necropsy
- Target organ weight
- Target organ histology
- (clinical pathology)

Repeat dose toxicity

- Normally preceded by Dose Range Finding study
- 2 species - rodent + non-rodent
- Duration of studies dependent on duration of human treatment
- Toxicokinetics required to document exposure and aid to interpretation
- Recovery period to investigate reversibility of findings

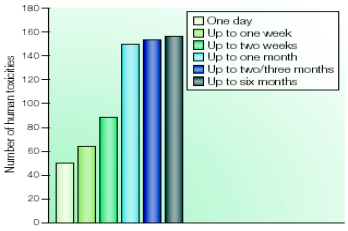


Figure 5 | **Time to first detection of animal toxicity.** The number of toxicities that can be detected in animal systems reaches a plateau at the one-month stage of the study. By this time, 94% of toxicities were detected, but prior to this time some toxicities were not apparent. On the first day, 25% of these observations were from safety pharmacology rather than from toxicology studies. Modified, with permission, from REF. 12 © (2002) Elsevier Science.

Role of toxicological risk assessment

- Due to a new chemical synthesis of the drug substance to be used in clinical trials a new impurity is found in the drug product:
  - Question to the toxicologist: Can we use this batch of drug product in clinical trials
- During migration studies using a new type of plastic bottle a new impurity is observed in a marketed drug product
  - Question to the toxicologist: can we release this product in this new bottle to the market.

Toxicological risk assessment in pharmaceutical industry

- Perform new toxicology studies to qualify the impurity
- Study own documentation: was this impurity present in batches used in safety testing
- Perform literature study and try to find No-effect levels

Permitted Daily Exposure

- PDE (mg/day) = $\frac{\text{NOEL or LOEL (mg/kg) x human body weight (50 kg)}}{\text{F1 x F2 x F3 x F4 x F5}}$

- F1: Interspecies differences,
  - mouse:human = 12
- F2: Inter-individual differences
  - 10
- F3: Duration of exposure
  - 10 short-term exposure
- F4: Severity of toxicity
- F5: Quality of data
  - 1 (NOEL determined)

Permitted Daily Exposure

- PDE (mg/day) = $\frac{\text{NOEL or LOEL (mg/kg) x human body weight (50 kg)}}{\text{F1 x F2 x F3 x F4 x F5}}$
- PDE = $\frac{\text{24 mg/kg x 50 kg}}{\text{12 x 10 x 10 x 1 x 1}}$ = 1 mg/day

Theoretical Maximum Exposure

- The theoretical maximum exposure [TME] to an impurity = maximal dose of drug product x concentration of the impurity in drug product.
- Risk assessment:
  - Divide Permitted Daily exposure with Total Maximum exposure and determine safety factor