

New challenges in the preclinical safety assessment of biopharmaceuticals

S2M Science to Market Conference
Vienna, 24-25 Feb 2010

Roy Forster
CIT, Evreux



EUROPEAN ASSOCIATION OF
PHARMA BIOTECHNOLOGY

Preclinical safety data

Phase I ('IND enabling') studies

- Preliminary studies
 - Single dose toxicity or DRF studies
- Repeated dose toxicity
 - 14 or 28 day studies in rodent & non-rodent species by clinical route. Test to maximum tolerated dose or other practical limitation. Should include toxicokinetic data, as preliminary indication of pharmacokinetic behaviour.
- Safety Pharmacology
 - Single dose *in vivo* studies to investigate potential secondary effects on CV, CNS and respiratory functions. *In vitro* electrophysiology study to assess potential for QT prolongation
- Genetic toxicology
 - *In vitro* studies for gene mutations and chromosomal aberrations.

Full development dossier

- Chronic toxicity studies
 - Two mammalian species
- Reproductive toxicity studies
 - Fertility, embryo-fetal toxicity etc
- Juvenile animal toxicity
- Pharmacokinetic and toxicokinetic data
 - Absorption, distribution, metabolism, excretion
- Carcinogenicity studies

Safety of biopharmaceuticals

EPO, neupogen and cancer patient survival

Eprex / PRCA incident

TGN1412 incident

TNF inhibitors and pediatric cancer

Tumour promotion of modified insulins

Tysabri and PML

small molecules vs biotech products

typical small molecule

Small molecules

Cost of goods low

Oral absorption

Hepatic metabolism

Active in wide range of species

Toxicity consequence of chemical structure

typical biotech product

High molecular weight

Cost of goods high

Parenteral administration

Degradation by plasma and tissue enzymes

Species restriction is common

Exaggerated pharmacology

ICH S6 guidance “Highlights”

INTERNATIONAL CONFERENCE ON HARMONIZATION OF TECHNICAL
REQUIREMENTS FOR REGISTRATION OF PHARMACEUTICALS FOR HUMAN USE

ICH HARMONISED TRI-PARTITE GUIDELINE

MAINTENANCE OF THE ICH GUIDELINE ON
NON-CLINICAL SAFETY STUDIES FOR THE CONDUCT OF
HUMAN CLINICAL TRIALS FOR PHARMACEUTICALS

Recommended for Adoption
at Step 4 of the ICH Process
on 10 July 1997 and
revised on 17 November 2000
by the ICH Steering Committee

This Guideline has been developed by the appropriate ICH Expert Working Group and
has been subject to consultation by the regulatory parties in accordance with the ICH
Process. At Step 4 of the Process the Guideline is recommended for adoption to the
regulatory bodies of the European Union, Japan and USA.

Selection of relevant animal model

- “in certain justified cases one relevant species may suffice”

Repeat dose toxicity

- “For biopharmaceuticals intended for chronic indications, studies of 6 months duration have generally been appropriate”

Immunogenicity

- “Antibody responses should be characterised (e.g., titer, number of responding animals, neutralising or non-neutralising) andcorrelated with any pharmacological and/or toxicological changes”

Reproductive toxicity studies

- The need for reproductive/developmental toxicity studies is dependent upon the product, clinical indication and intended patient population

Evaluation of carcinogenicity

- Standard carcinogenicity bioassays are generally inappropriate for biotechnology-derived pharmaceuticals. However, product-specific assessment of carcinogenic potential may still be needed.

What are the issues?

1. Selection of relevant animal model
2. (Role of tissue cross reactivity studies)
3. Immunogenicity
4. Reproductive toxicity studies
5. Evaluation of carcinogenicity

Relevant animal model

- “the selected species should be responsive to the primary pharmacodynamic effect of the substance”
 - “A relevant animal species is one in which the biopharmaceutical elicits a similar biological response as that expected to occur in humans, due to the expression of a responsive orthologous drug receptor/antigen”
- regulatory authorities expect (GLP) data demonstrating relevance
 - *in vivo* (eg. pharmacology), or *in vitro* (eg receptor binding)

species restriction

| | |
|------------------------------|-------------------|
| DNAase (Pulmozyme) | Not restricted |
| hGH (Protropin) | Not restricted |
| Engerix | Not restricted |
| IFN alpha2a (Roferon) | Restricted |
| IFN alpha2b (Intron) | Restricted |
| IFN beta (Avonex) | Restricted |
| rh-IL-2 (Proleukin) | Restricted |
| G-CSF (Neupogen, filgrastim) | Not restricted |
| GM-CSF (Molgramostim) | Not restricted |
| Erythropoietin (Eprex) | Not restricted |
| Trastazumab (Herceptin) | Restricted |
| ReoPro (Abciximab) | Restricted |
| Remicade (infliximab) | Restricted |
| Vitravene (fomivirsen) | Not restricted |

Relevant animal model

Conventional safety evaluation often problematic.

- In some cases there are no animal models that have the relevant molecular target, receptor or epitope.

Is it useful to perform toxicology studies in non-responsive animal species?

- ICH S6 states that “Toxicity studies in non-relevant species may be misleading and are discouraged”. FDA pharm/tox reviews generally disqualify studies in non-relevant species (“not relevant to the evaluation of safety ...”)

Safety studies using homologous (surrogate) products:

- Approach used for Actimmune (interferon-gamma), Remicade (Infliximab), Raptiva (efalizumab), Cimzia (Certolizumab) and Solaris (eculizumab)

Safety studies using humanised transgenic model:

- Example: keliximab (anti-CD4 monoclonal)

.....new proposals

Proposed (Addendum to ICH S6)

1. For monoclonal antibodies ...directed at foreign targets it is desirable to evaluate safety in an animal model of disease.
2. If there are two pharmacologically relevant species, both should be used for short term studies (eg 28 days). If the findings are similar, one species can be sufficient for longer term studies.
3. Rationale for high dose selection based on whichever is higher: dose inducing maximum pharmacological effect or 10-fold clinical exposure

immunogenicity

Previous

- “Antibody responses should be characterised (e.g., titer, number of responding animals, neutralising or non-neutralising) andcorrelated with any pharmacological and/or toxicological changes”
- An antibody response should not trigger early termination of a preclinical toxicology study unless the pharmacological / toxicological effects are neutralised in a large proportion of animals. If interpretation is not compromised by these issues, no special importance should be ascribed to the antibody response
- The induction of antibody formation in animals is not predictive of a potential for antibody formation in humans

immunogenicity

Proposed

Blood samples should be taken but in the absence of PK or PD changes, determination of anti-drug antibody levels is not needed.

“Conventional safety evaluation often problematic”

Reproductive toxicity

Segment I : effects on male and female fertility (rat)

Segment II : embryo-fetal toxicity, EFD (rat and rabbit)

Segment III : peri- and post-natal study, PPND (rat)

Juvenile toxicity (rat and other species)

Carcinogenicity

Two year rodent bioassay (rat and mouse)

reproductive toxicity

- Reproductive toxicity studies should be conducted only in pharmacologically relevant species
- Studies in NHP are preferred over approaches using alternatives (surrogate, homologous)
 - NHP are not good models for fertility trials. Fertility can be assessed through histopathology (in M+F) and cyclicity (in F)
 - For embryo-fetal development (EFD) studies, placental transfer of the biopharmaceutical drug should be taken into account
 - A single well-designed study can cover EFD and PPND. ICH indicates the statistical power required.

reproductive toxicology

- Primate studies
 - difficult to source purpose-bred sexually mature animals
 - long (long gestation, period to weaning)
 - expensive
 - low litter size (one fetus per pregnancy), hence large groups sizes and/or low statistical power
 - high abortion rate of primates in capture
- overall, a hard choice

Example EFD + PPND design

- 20 pregnant females per group
- Control group and 2 treatment-levels
- Administration of test item during period of organogenesis (eg 10 administrations over GD20 to 50)
- Samples for TK, ADA, FACS analyses etc
- Cesarean section (10 animals per group) at GD 100 (metrics, morphology, soft tissue and skeletal examinations)
- Delivery at term, GD 160 to 170 (10 animals per group); developmental metrics, landmarks of functional development; follow-up through 12 months

placental transfer

Placental transfer of antibodies in humans

- Placental transfer of IgG is mediated by an Fc-receptor
 - Month 3: Maternal IgG appears in fetal circulation
 - Month 8: Fetal IgG levels are 50% of maternal levels
 - At birth: Fetal IgG levels equal or greater than maternal
- FcRn receptors on syncytiotrophoblast and on intestinal epithelium
- (Fc receptors are also involved in regulating the plasma half-life of antibodies)

transfer of maternal antibodies

| | Placental transfer | Colostrum |
|------------|---------------------------|----------------------|
| Human | 100% | |
| Primates | 100% (IgG) | 0 |
| Rabbit | 100% (IgG, IgM) | 0 |
| Rodents | 10% (IgG) | 90% (IgG, IgA, IgM) |
| Dog, cat | 5% (IgG) | 95% |
| Pig, sheep | 0 | 100% (IgG, IgA, IgM) |

segment II studies

- Selection of species to take account of:
 - Relevant receptor or epitope
 - Placental passage of antibody
- Frequency of administration to take account of:
 - clinical administration
 - immunogenicity
 - critical periods in development

carcinogenicity

- Can a protein be carcinogenic?
 - PTH 1-34 induction of osteosarcomas
 - Modified insulin AspB10 induction of mammary gland adenocarcinomas in rats
 - EPO and tumour growth
- Particular concern about growth factors and immunosuppressants

carcinogenicity

- Rodent studies may be precluded because of species restriction and/or immunogenicity and/or route of administration.
- Product-specific assessment of carcinogenic potential may still be needed
- Complex decision tree proposed; in general product specific strategies preferred to rodent bioassays

carcinogenicity

- Conventional rodent assay
 - Abatacept : mouse subcutaneous (sc) carcinogenicity study
 - Insulin glargine (Lantus): rat and mouse sc carcinogenicity studies
 - Mecasermin, r-hu-IGF1 : rat sc carcinogenicity study

- Conventional rodent assay with homologous protein
 - recombinant mouse growth hormone: mouse sc carcinogenicity study
 - recombinant rat growth hormone: rat sc carcinogenicity study

carcinogenicity

- Short term transgenic assays
 - PegIntron (peginterferon alfa-2-b); 26 week carcinogenicity study in p53^{+/-} mice
 - Mouse homologue of efalizumab (Raptiva); 26 week carcinogenicity study in p53^{+/-} mice
- Evaluation of tumour growth in xenograft models expressing relevant receptors
 - Insulin glulisine

carcinogenicity

Indirect data

- Infliximab (monoclonal specific for human TNF α), Carcinogenicity was evaluated using two models:
 - TNF α knockout mice
 - The effect of anti-murine TNF α antibodies on tumour development in the mouse

conclusions

- Protein therapeutics, monoclonal antibodies and biopharmaceuticals in general are under increased scrutiny
- Some new approaches will facilitate nonclinical development
- Other requirements bring significant challenges
- Thank you for your attention, enjoy your lunch!