Alternative Methods: The Challenge of New Technologies

H. J. Ahr, Wuppertal
Bayer HealthCare AG, Special Toxicology
Brussels, November 24-25, 2007

Alternatives in Toxicology

Alternative methods – Safety first!

- Industry fulfils expectations of the society for chemical products (pharmaceuticals, crop protection, chemicals...)
- The protection of customers, population and environment is of highest priority. Traditionally animal tests are used for safety assessment.
- Where ever possible, safety evaluation should be accomplished by a minimum of animal experiments (3 R Principle) but without compromising the level of safety.

3R: Refine
Reduce
Replace

Alternatives in Toxicology

We have been not very successful in realizing the 3R principle!

Area	**Replacement by *in vitro* alternative**	
Acute toxicity	not foreseeable	( )
Toxicity after multiple dosing	not foreseeable	( )
Reproductive toxicity	not foreseeable	( )
Genotoxicity	partially implemented	( )
Carcinogenicity	not foreseeable	( )
Local tolerance	possible, if validation successful	( ) !
ADME	not foreseeable	( )

CPMP 1997
(CSTEE 2004)

Alternatives in Toxicology

What's the consequence?

Conventional testing in animals will continue until properly validated and accepted alternative methods will be available

- to guarantee safety according to the state of the art

- to avoid liabilities

- to follow international regulations (not only Europe!)

Up to now, Refinement and Reduction are the preferred options to reduce animal numbers

Alternatives in Toxicology

May be we expected too much!

Major scientific hurdles exist (amongst others):

- The issue of complexity: an organism is more than the sum of its cells
- The in vitro models currently available are largely insufficient and inable to mirror in vivo effects
- To simulate an intact organism by simply cumulating in vitro tests for organ toxicity is the wrong way for scientific and practical reasons
- Our current paradigm to develop alternative methods thus is obviously insufficient

Alternatives in Toxicology

The issue of complexity

Animal models

- complex
- cell-cell and systemic interactions
- metabolism and toxicokinetics represented

- multiple endpoints / organs
- reversibility and regeneration

- chronic effects
- years of experience in risk assessment

In vitro models

- reductionistic (single cells, mechanisms)
- lack of complex interactions
- influence of metabolism and toxicokinetics difficult to simulate

- not available for many endpoints / organs

- correlation to defined toxicological phenotypes difficult
- short time models
- hazard

Alternatives in Toxicology

The issue of complexity is not (yet?) solved…

… even for low molecular weight compounds

- In vitro models are only in place for isolated specific questions
- Replacement of even simple animal tests will need test batteries of multiple vitro models, if possible at all

But

- The knowledge of the complex interactions is still largely lacking
- Consequently, the extrapolation to in vivo animal/human is uncertain
- No such test battery has ever been validated
- Cost of such test batteries may be critical

Need for new thinking

Alternatives in Toxicology

Elements of a novel approach based on new technologies

- Mechanistic understanding of toxicity fueled by new technologies (omics, transgenics, …)
- Mechanism based in vitro tests (not organotypic models)
- New ways to describe the complexity (PBPK, systems biology)
- Economically feasible test batteries (High content screening, HTS technologies)

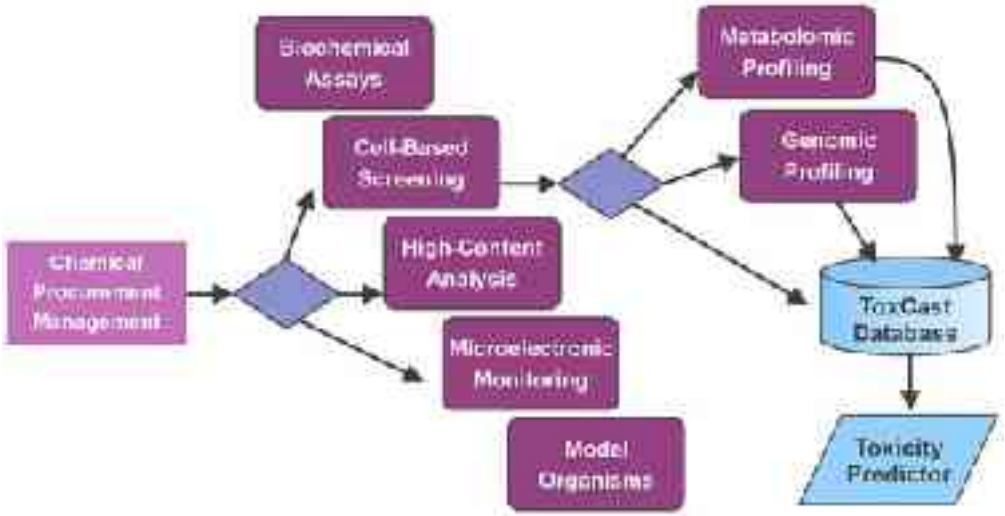
and… and…and…

a lot of basic research

Alternatives in Toxicology

A first step in this direction

EPA's ToxCast™ Program for prioritizing toxicity testing



www.epa.gov/comptox/toxcast

Alternatives in Toxicology

New technologies will now add even more complexity

- New human specific targets for pharmaceuticals (genomics)
- Biotechnology derived pharmaceuticals (biopharmaceutical i.e. antibodies, proteins, peptides)
- Nanomaterials
- GMO's

⇒ Value of traditional testing paradigms in animals questionable

Alternatives in vitro not yet available

Alternatives in Toxicology

Biopharmaceuticals

Major challenge: Relevance of animal model:

Example: “TGN 1412 – a tragic accident, which raises numerous questions”

> *“unclear why the comprehensive preclinical investigations in non human primates gave no indication of the potentially lethal pharmacodynamic properties of TGN 1412, which become obvious in man”*
>
> B. Liedert et. al.
> J Clin Pharmacol (2007)

> *“Novel* (in vitro) *procedures would have predicted the toxicity of this super agonist”*
>
> R. Stebbings et. al.
> J Immunol (2007)

Alternatives in Toxicology

Biopharmaceuticals

Consequence of TGN 1412 – New guideline (2007):

"Toxicity studies in non relevant species may give rise to misinterpretations and are discouraged."

Relevant species: *"A weight of evidence approach should involve integration from in vivo, ex vivo and in vitro studies."*

EMEA 2007

⇒ **An integrated approach of relevant in vivo and specific in vitro tests is required.**

A new chance for alternatives?

Alternatives in Toxicology

Biopharmaceuticals

A new chance

Due to the diversity of biopharmaceuticals, a case by case approach
not a fixed testing scheme is proposed

Consist of in vivo and in vitro elements in relevant test models

Open for new technologies
(transgenics, omics, human cells ,homologous proteins)

⇒ Such intelligent test strategies may also provide learning cases for the testing of conventional products

Alternatives in Toxicology

The advent of new technologies and their products creates new challenges

but

May also contribute to new solutions