Claudius Griesinger, Thomas Hartung, ECVAM team.

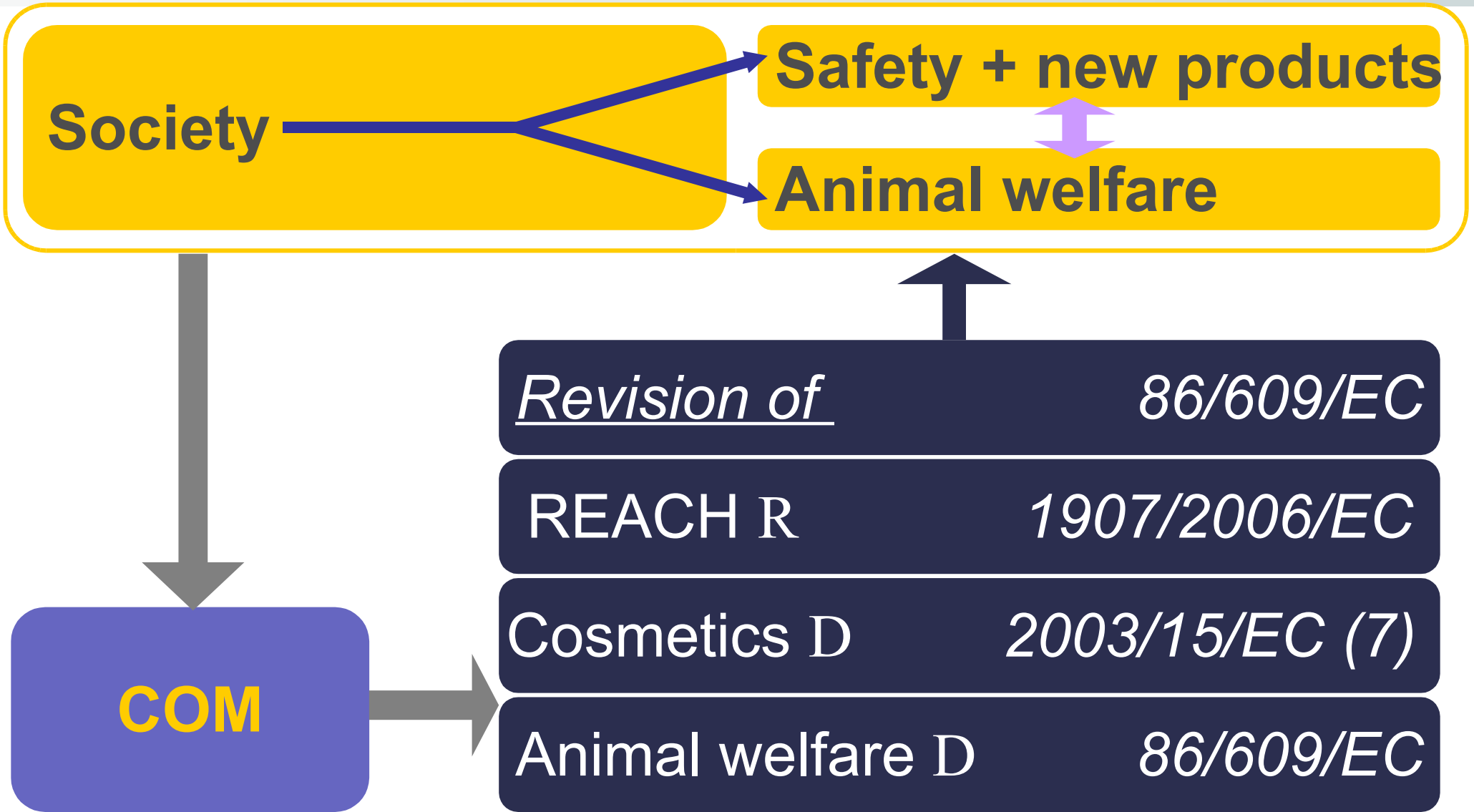
Key area leader

- *Evidence-Based Toxicology (EBT)*
- *CORRELATE ('Commission Reference Laboratory for Alternative Test Evaluation')*

OVERVIEW

- *Driving forces:*
  translating societal expectations into legislation
- *Key legislations:*
  REACH & Article XI
- *The "decade of toxicology"? Are we ready ?*
  Evidence-based Toxicology
- *Independent assessment of alternative methods*
  CORRELATE

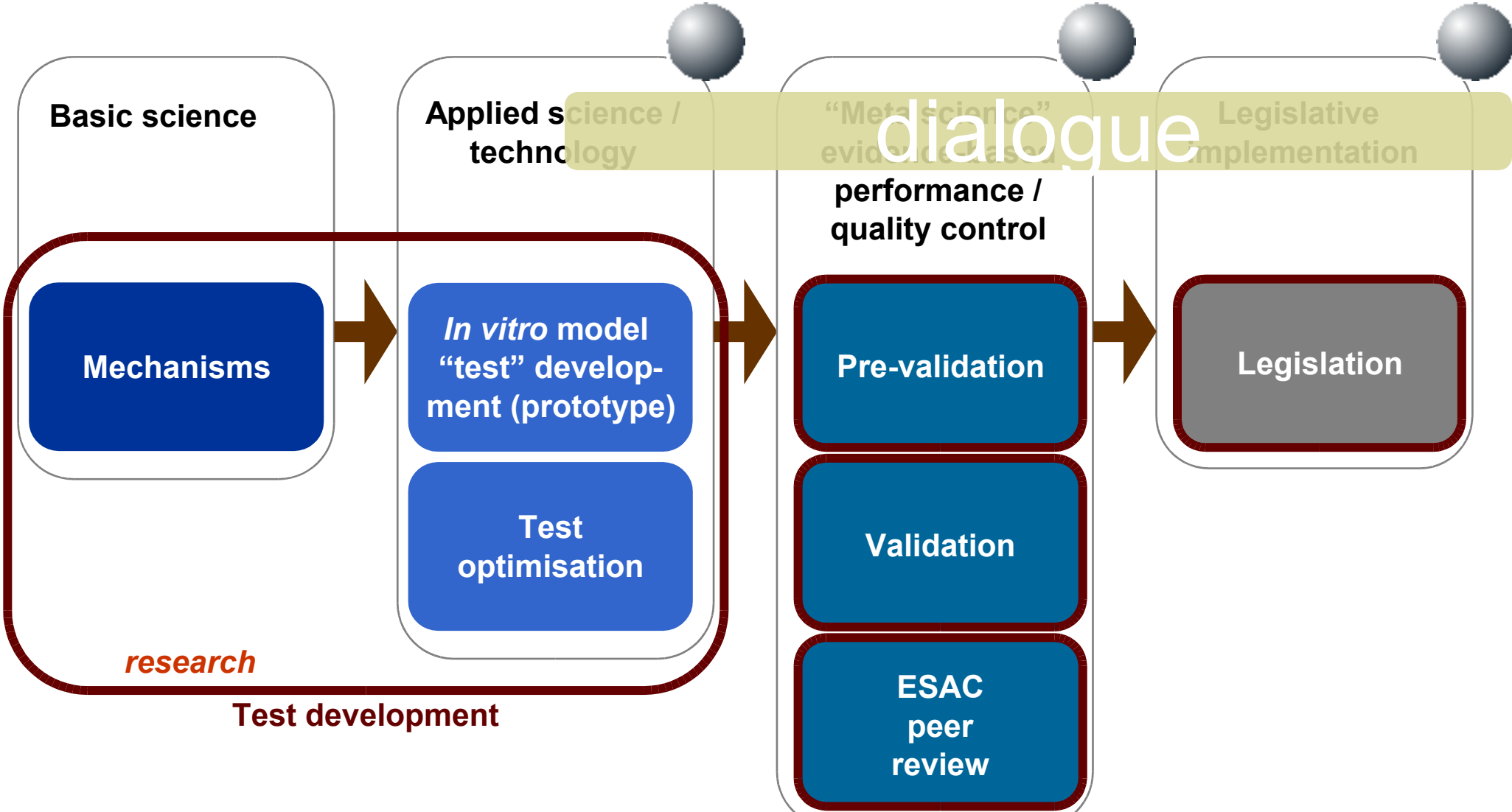
The driving force: societal expectations of sustainability



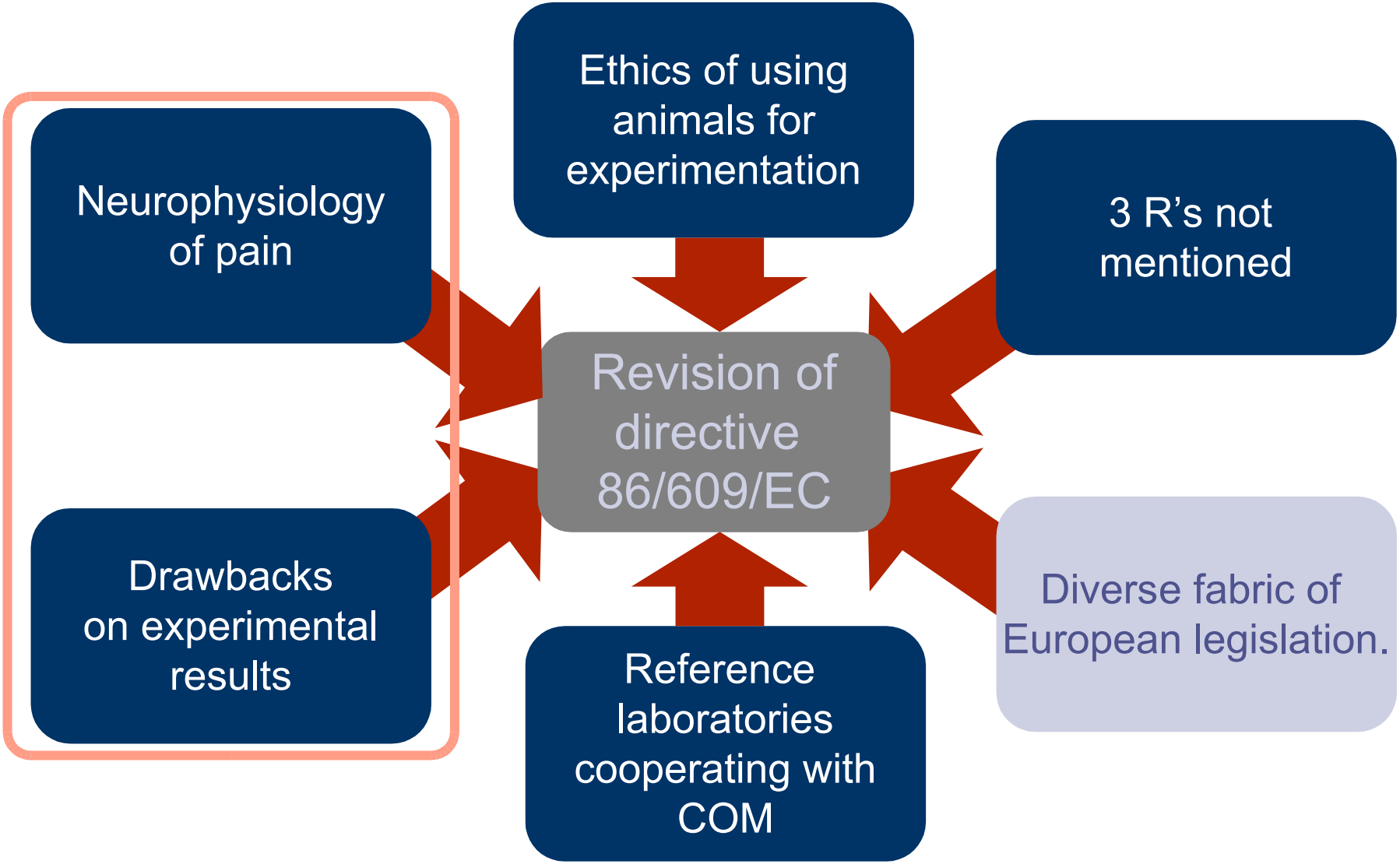
ESAC statements. *chemicals*

ESAC Statement	Date	67/548/EEC	OECD
• 3T3 NRU phototoxicity test	1997	2000	2002
• EPISKIN™ skin corrosivity test	1998	2000	2002
• Rat TER skin corrosivity test	1998	2000	2002
• 3T3 NRU phototoxicity test to UV filter chemicals	2000		
• Local Lymph Node assay for skin sensitisation	2000		2002
• EpiDerm™ skin corrosivity test	2000	2000	2002
• CORROSITEX ® skin corrosivity test	2000		2006
• Embryonic stem cell test for embryotoxicity	2002		
• Whole-embryo culture test for embryotoxicity	2002		
• Micromass test for embryotoxicity	2002		
• Micronucleus test, alternative to in vitro chromosome abberation	2006		
• SkinEthic skin corrosivity test	2006		
• BCOP / ICE test for identifying severe eye irritants	2007		
• rLLNA (reduced local lymph node assay – skin sensitisation)	2007		
• EPISKIN (MTT.IL1a) + EpiDerm (MTT) – skin irritation (full & partial replacement)	2007		

Ante / pre / validation / post



Revision of 86/609/EC



OVERVIEW

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  translating societal expectations into legislation
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REACH and alternative methods

Whereas 1

Article 1

Article 13

Article 25

Annex XI

REACH and in vitro – Whereas 1

Whereas 1

The purpose of this Regulation is to ensure a high level of protection of human health and the environment,
as well as the free movement of substances, on their own, in preparations and in articles while enhancing competitiveness and innovation. This Regulation should also promote the **development of alternative methods for the assessment of hazards of substances.**

REACH and in vitro – Article 1

Article 1

Aim and Scope

The purpose of this Regulation is to ensure a high level of protection of human health and the environment, **including the promotion of alternative methods for assessment of hazards of substances**, as well as the free circulation of substances on the internal market while enhancing competitiveness and innovation.

REACH and in vitro – Article 13

Article 13

General requirements for generation of information on i. p. of substances

(3)
To generate information, conduct in accordance with „...**test methods laid down in a Commission regulation** or other international test methods recognised by the Commission or the Agency as being appropriate.“

„....information ... may be generated in accordance with other test methods provided that the conditions set out in Annex XI are met.“

REACH and in vitro – Article 25

Article 25

Objectives and General Rules

In order to avoid animal testing, testing on vertebrate animals for the purposes of this Regulation shall be undertaken **only as a last resort.** It is also necessary to take measures limiting duplication of other tests.

REACH and alternative methods

Whereas 1	*Development*	
Article 1	*Promote their use in hazard testing*	
Article 13	*Not only 'canonized' methods -> val. AM's*	Annex XI
Article 25	*Their use indirectly encouraged*	

REACH – Annex XI

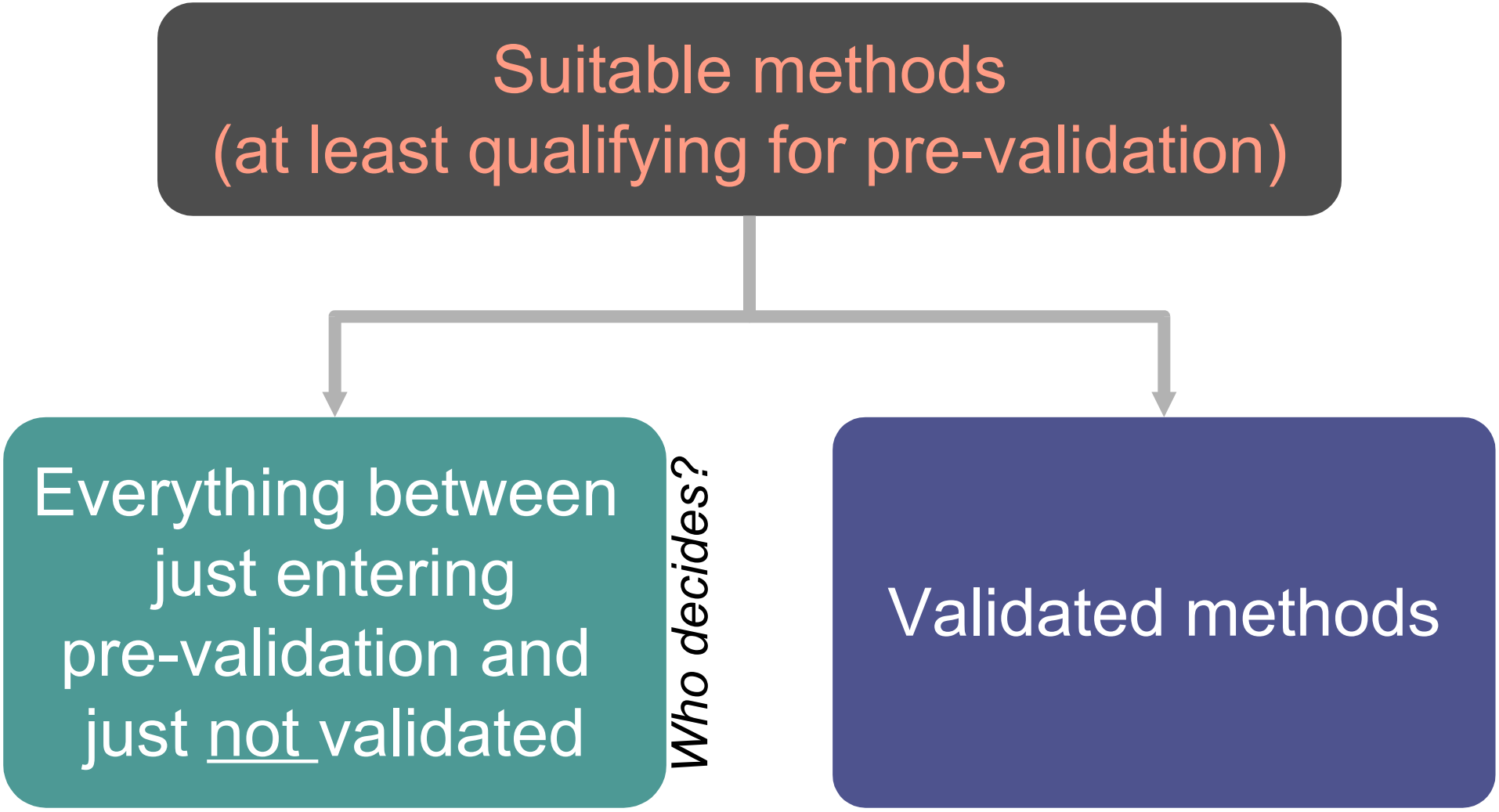
Annex XI

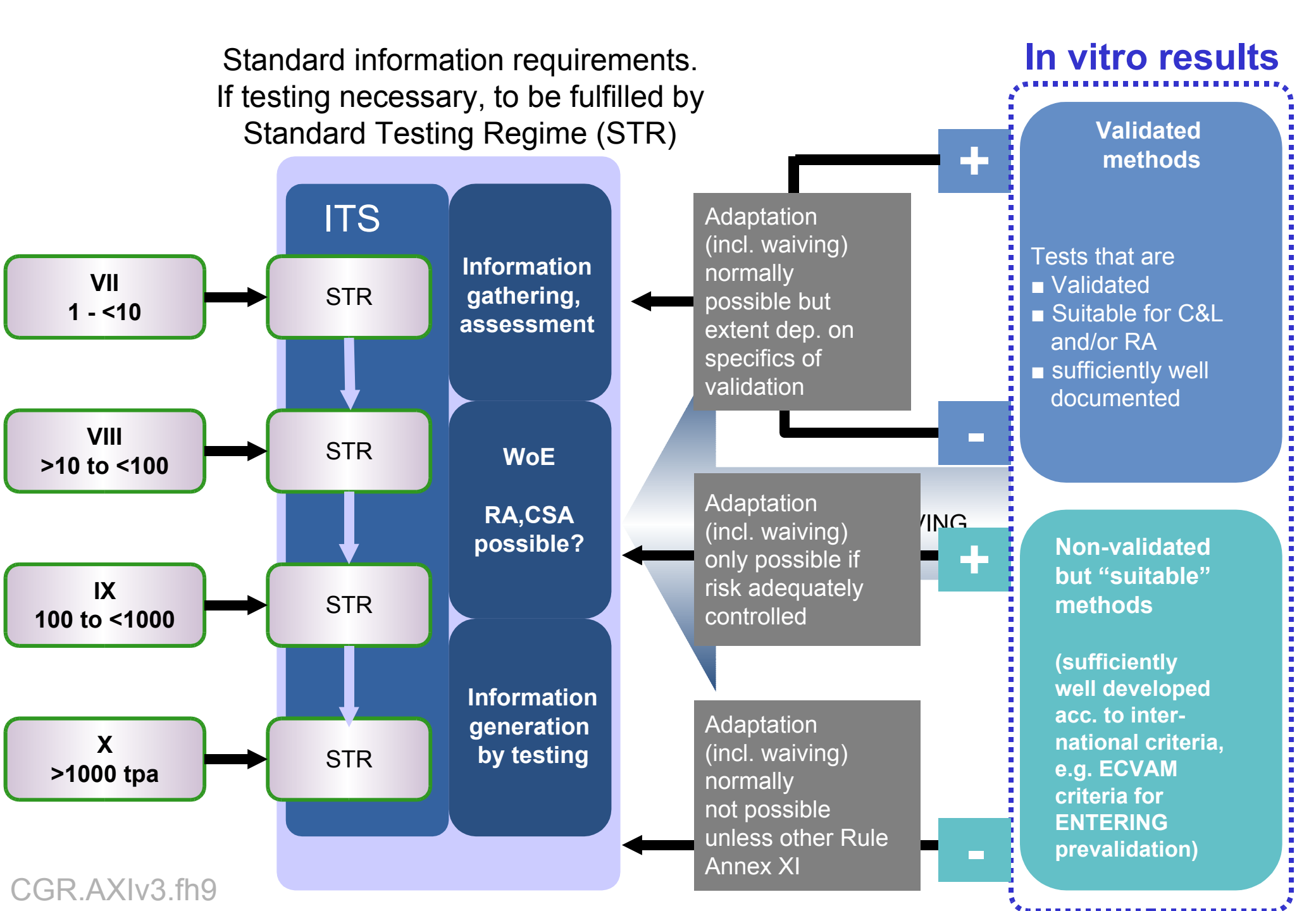
General rules for adaptation of the standard testing regime set out in Annexes VII to X

1. Standard (often Animal) **Testing does not appear scientifically necessary**

1.1 Use of existing data
1.2 Weight of evidence
1.3 Qualitative or quantitative structure-activity relationships
1.4 In vitro methods
1.5 Grouping of substances and read-across approach

Suitable methods according to REACH





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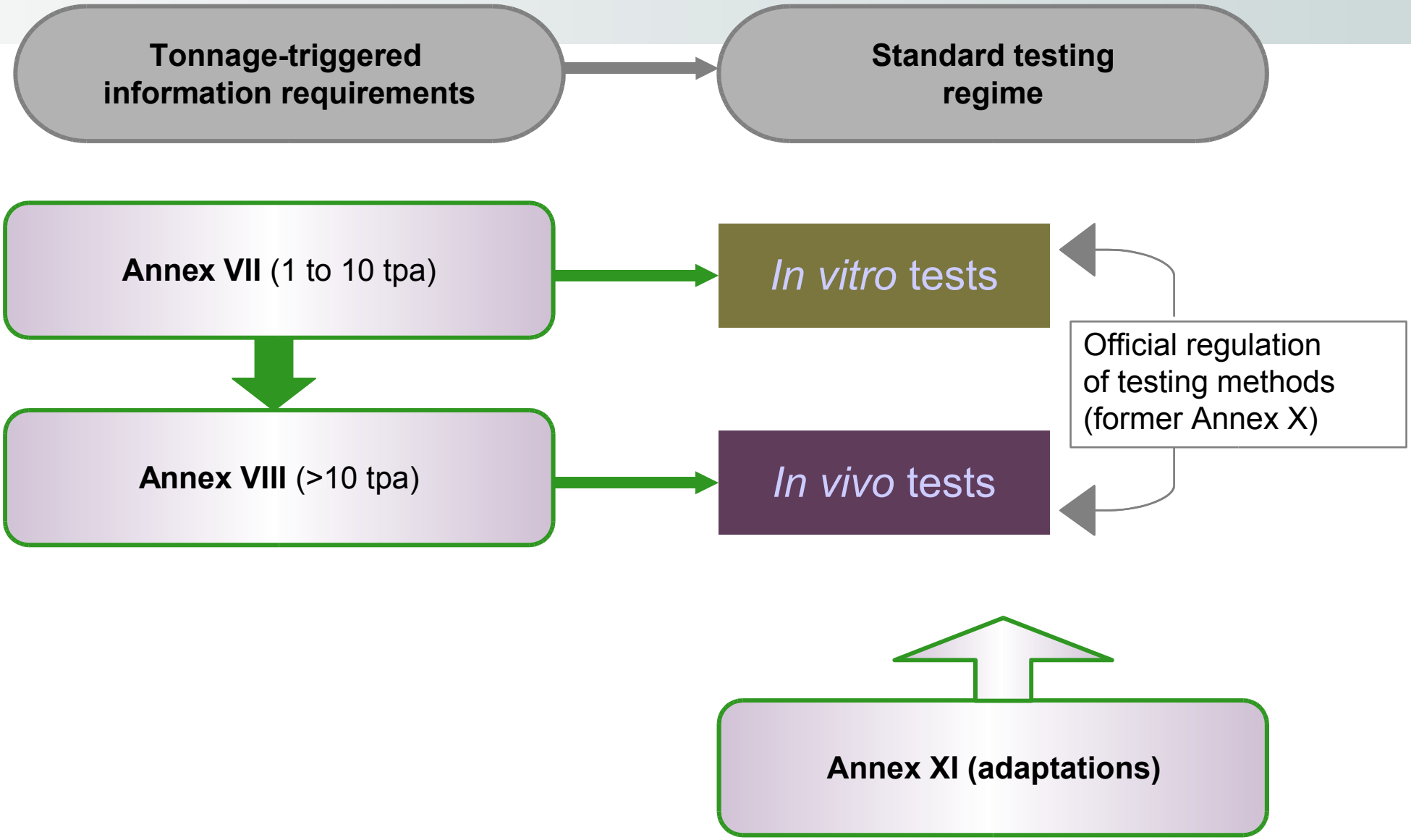
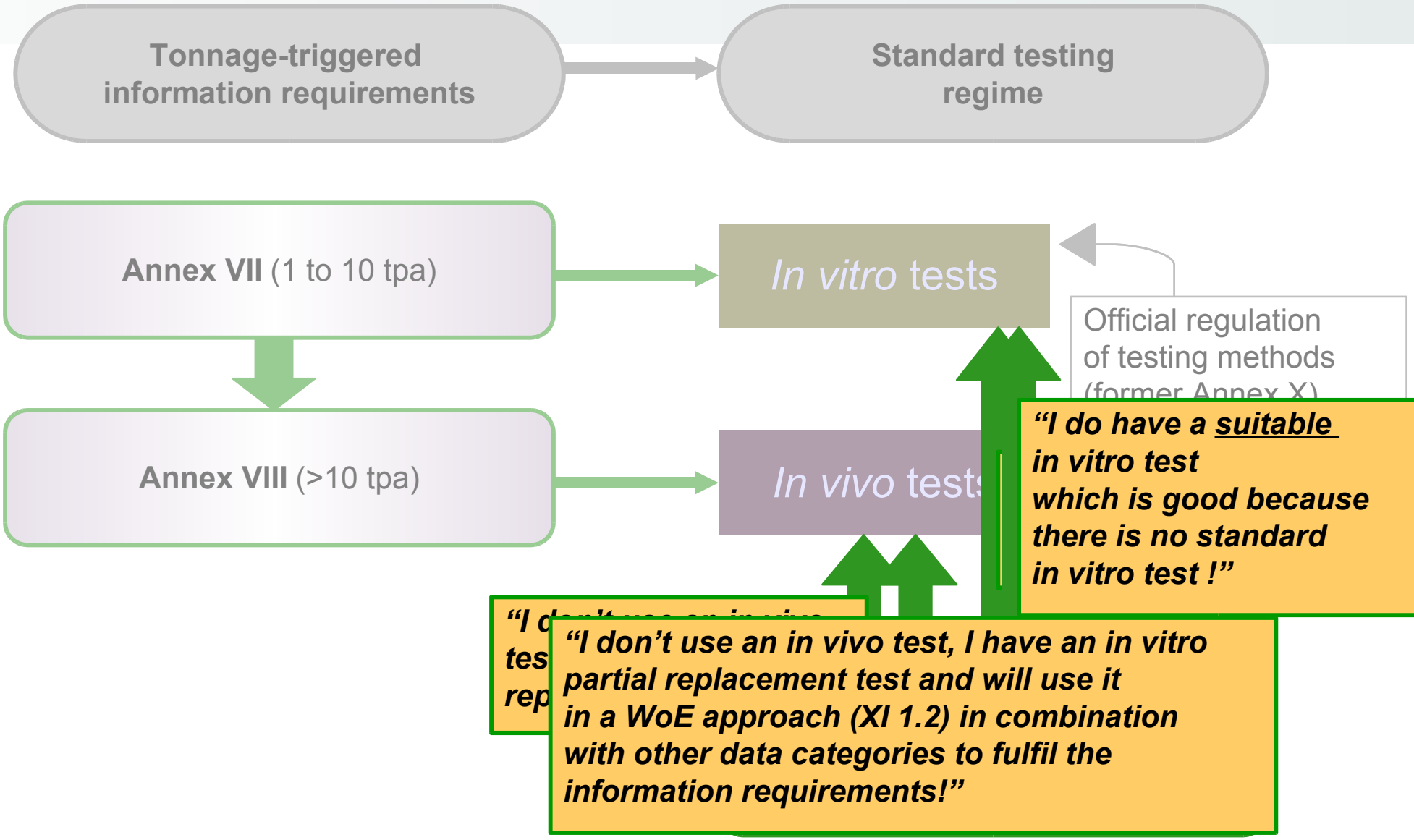

Tonnage-triggered information requirements
Standard testing regime
Annex VII (1 to 10 tpa)
Annex VIII (>10 tpa)
In vitro tests
In vivo tests
Official regulation of testing methods (former Annex X)
Annex XI (adaptations)


Tonnage-triggered information requirements
Standard testing regime
Annex VII (1 to 10 tpa)
Annex VIII (>10 tpa)
In vitro tests
In vivo tests
Official regulation of testing methods (former Annex X)
"I do have a suitable in vitro test which is good because there is no standard in vitro test !"
"I don't use an in vivo test, I have an in vitro partial replacement test and will use it in a WoE approach (XI 1.2) in combination with other data categories to fulfil the information requirements!"

RIP 3.3 ECVAM's contribution

Stakeholder expert group (SEG) input process:
Thomas Hartung, Christoph Klein

Contribution to "endpoint working groups" (EWGs):

- Acute toxicity (*Laura Gribaldo*)
- Sensitisation (*Costanza Rovida*)
- Reproductive toxicity (*Susanne Bremer*)
- Toxicokinetics (*Michel Bouvier d'Ivoire*)
- Mutagenicity, Carcinogenicity (*Raffaela Corvi*)
- Environmental/aquatic toxicity (*Marlies Halder*)
- Skin & eye corrosion/irritation & respiratory irritation.

(*Chaired by ECVAM*: *Valerie Zuang, Claudius Griesinger*).

Drafting group RIP 3.3
Christoph Klein, Costanza Rovida, Claudius Griesinger

OVERVIEW

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  CORRELATE

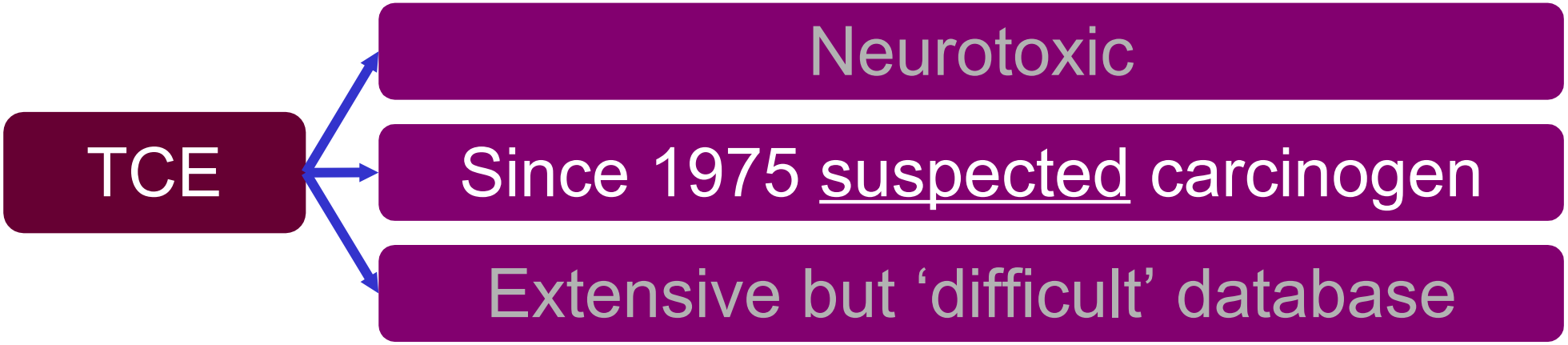
Evidence-based toxicology

Toxicology is a success story, but there is some room for improvement !

- Lack of adaptation to progress in life & toxicological sciences
- Traditional assessment methods: Unknown reliability & relevance
- Decision-making on risks & hazards: No global “best practice”, consensus-driven, en-route criteria
- Data integration – lack of quantitative and more objective methodologies
- No mechanisms to listen to societal expectations (sustainability)

Example: Risk assessment of trichloroethylene

29 risk assessments (animal & human data) analysed



Rudén C. The use and evaluation of primary data in 29 trichloroethylene carcinogen risk assessments. *Regul. Toxicol Pharmacol 2001; 34: 3-16.*

Example: Risk assessments of trichloroethylene

Carcinogen 4 studies | **Equivocal** 19 studies | **No carcinogen** 6 studies

- average reference coverage **18%**
- average citation coverage of most relevant studies **80%**

} selection bias

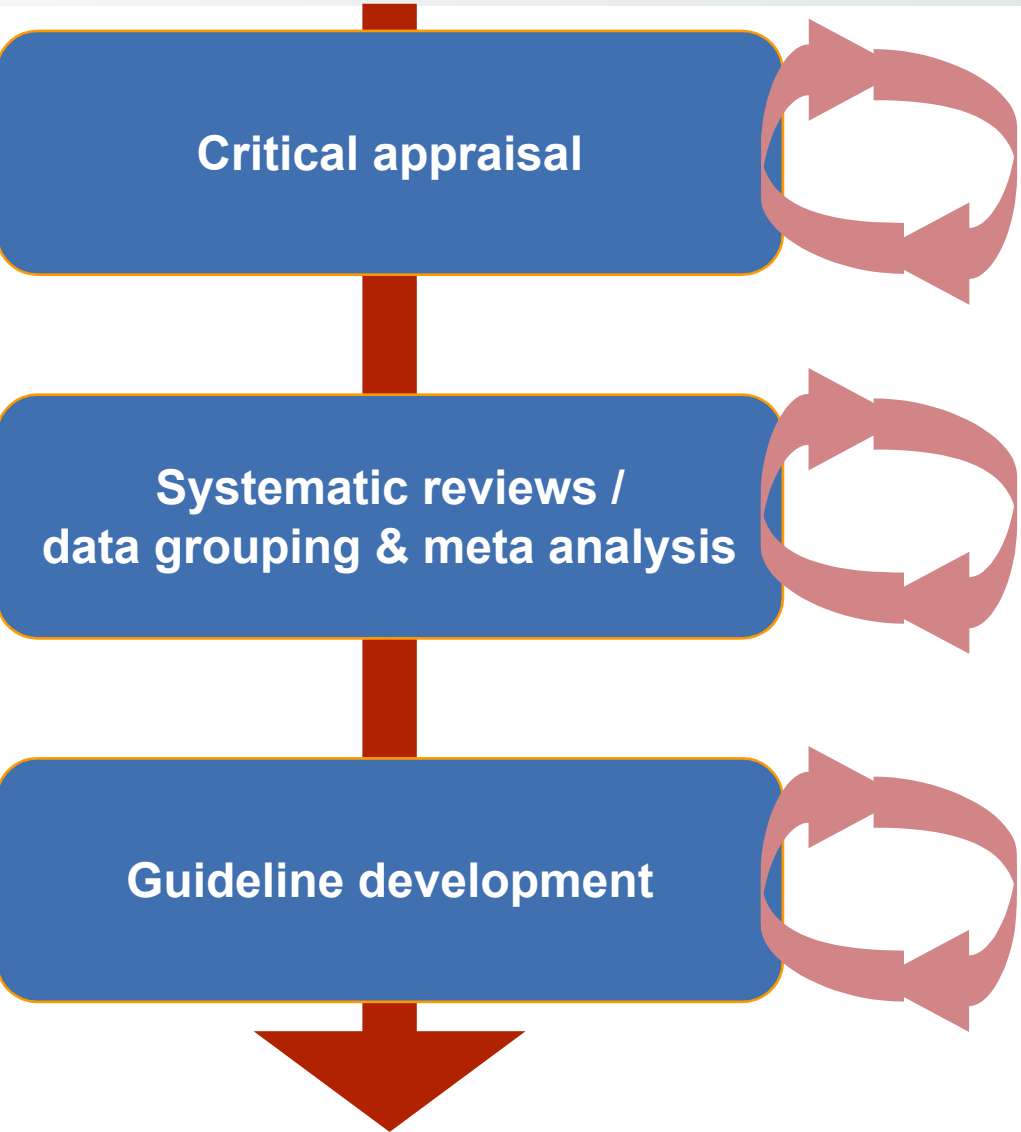
- interpretation differences of most relevant studies in **27%**
- study/data quality: assessment not documented in **65%**

Reasons for differences in risk assessments

⇒ bias in data selection (incomplete and diverse)

⇒ different data interpretation/evaluation

Evidence based medicine - tools



- Continuous adaptation to scientific progress.
- Transparency.
- Explicitness.

- Conscientious, Judicious use of best "evidence".

Evidence-Based Medicine: systematic reviews

Question — How much more reliable/efficient is diagnostic method X compared to Y detect Z

Standards — Define *ex ante* criteria for **search**, **inclusion**, **evidential power** of information

Minimising intrinsic bias

"Protocol"
(Study plan, review plan)

- Question
- Standards

Peer review (I)

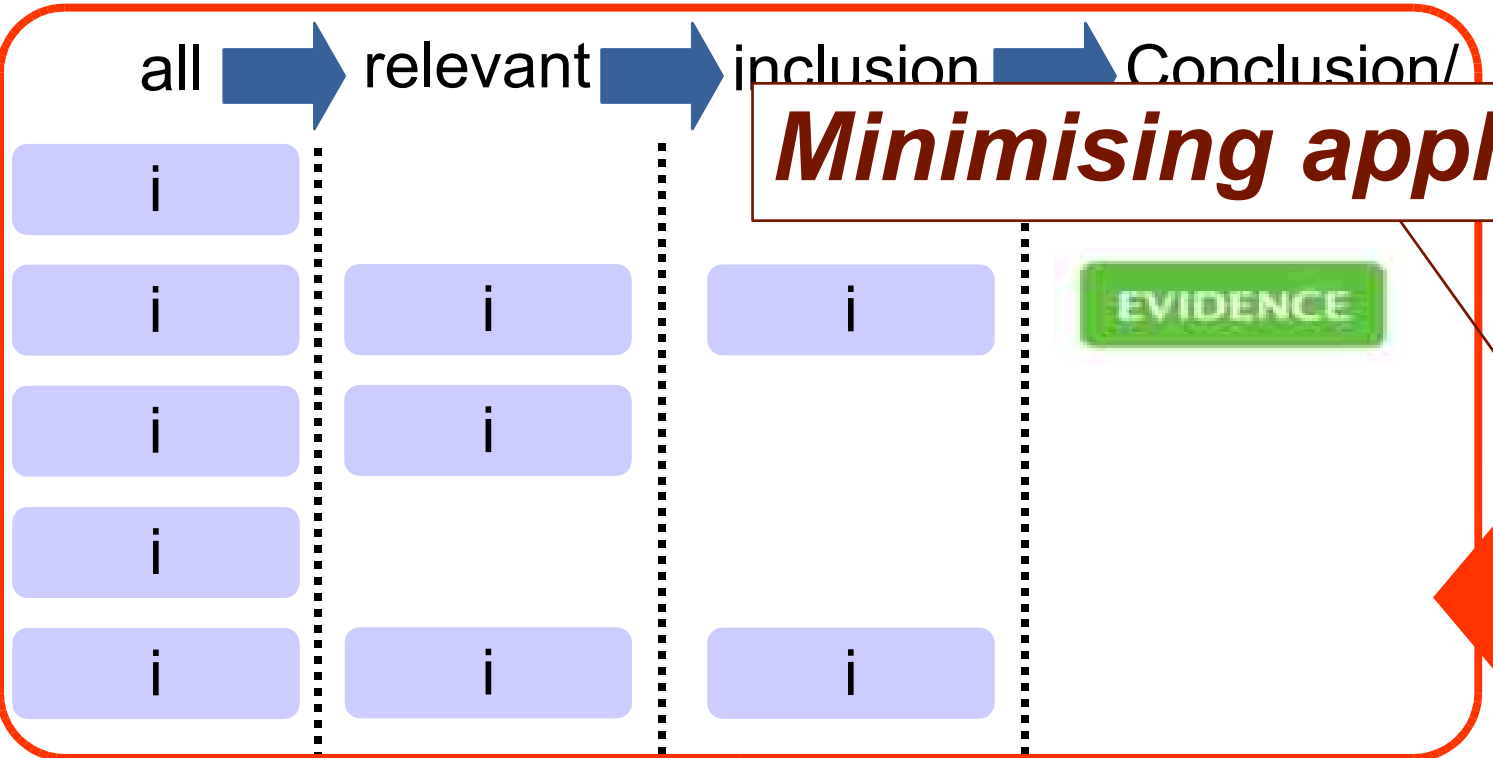
Evidence-Based Medicine: systematic reviews

Question

How much more reliable/efficient is diagnostic method X compared to Y detect Z

Standards

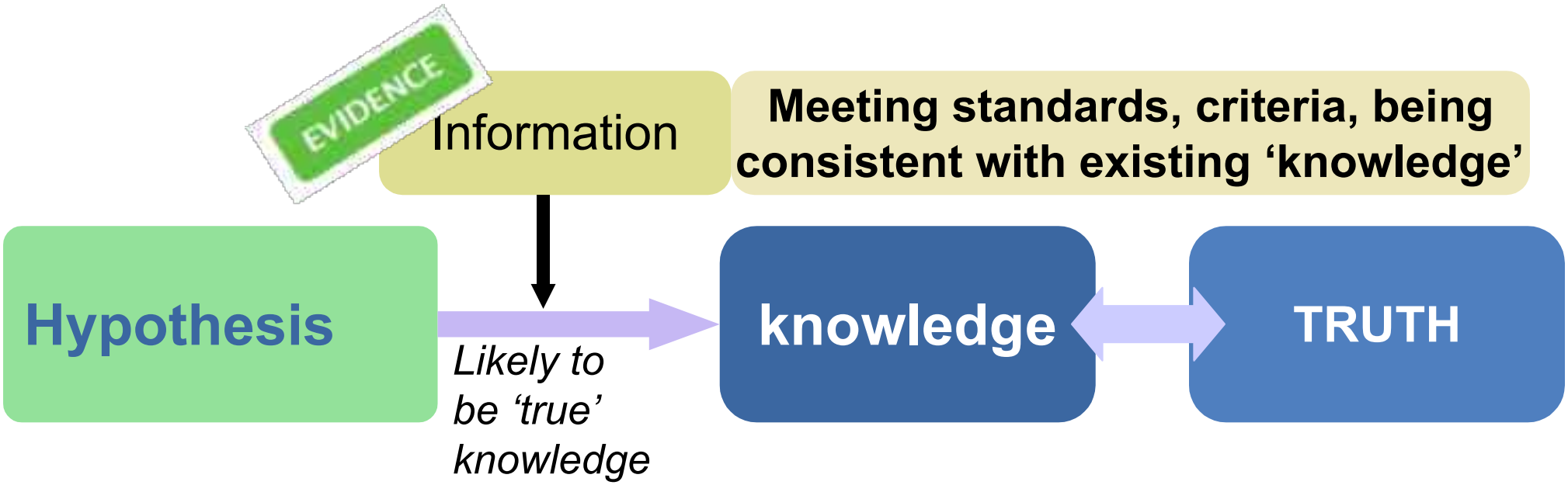
Define ex ante criteria for **search**, **inclusion**, **evidential power** of information



Minimising application bias

Peer review (II)

What actually is evidence ? – epistemology



- *What **criteria / standards** must **information** fulfil to be regarded as **evidence** for / against a hypothesis so that we can regard the hypothesis as **probable knowledge?***

1st International Forum towards Evidence-Based Toxicology (EBT)

EBT

15-18 October 2007, Como, Italy

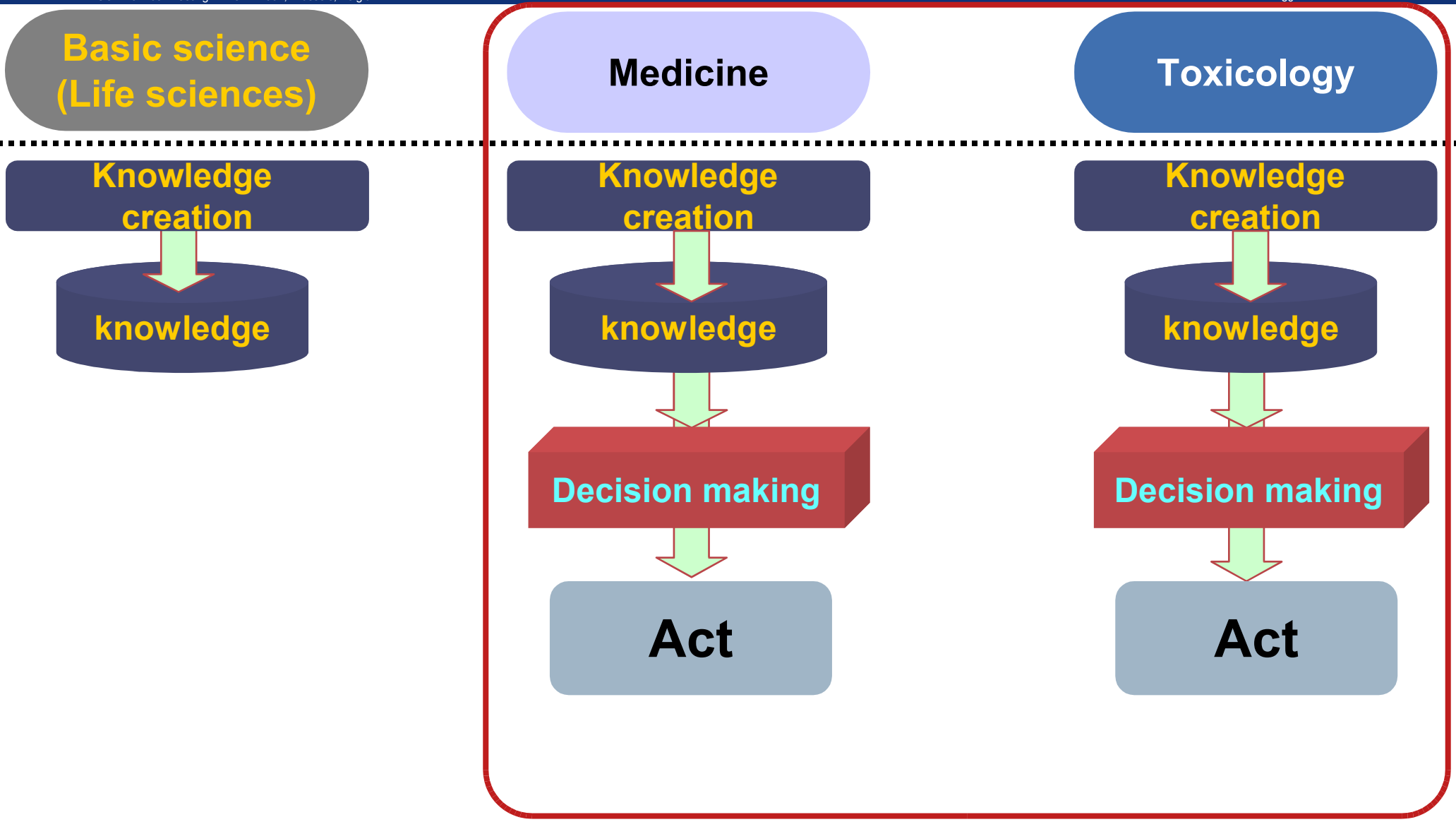
www.ebtox.org

About 170 participants

- From basic research, industry, regulation, animal welfare, policy making…
- From > 25 countries
- From Europe, Africa, Asia, America

Two of the core questions of the forum

- *Kaizen* : How can we further improve toxicological practice ?
- **Can toxicology profit from evidence-based approaches (e.g. EBM) ?**
- **How can we used Evidence-Based tools in Toxicology (EBT) ?**
- How do we define EBT, what are its objective and aims ?
- How can we make EBT workable ?



A craft (e.g. metallurgy)

Medicine: causation & probability

Medicine

Hypotheses on causal links (causation)

- ***Z*** ⟶ ***A*** (adverse effect)
- ***C,P*** ⟶ ***D*** (desired effect), ***A*↓**

Probability of ***A*** to **occur** (probability)
Probability of ***C*** to cause add. adverse effects ***A2***

Probability of ***C*** (cure) to reduce ***A*** (probability)
Probability of ***P*** (prevention) to avoid ***A*** (probability)

Disease, hazard

Pathogenesis

Curative/preventive acts

Risk

Effectiveness

Toxicology: causation & probability

Toxicology

Hypotheses on causal links (causation)

- ***Y*** ⟶ ***A*** (adverse effect)
- ***P*** ⟶ ***A*↓**

Probability of the ***A*** to occur (probability)

Probability of the ***P*** (RRM) to prevent ***A*** (probability)

Hazard

"Toxicogenesis"

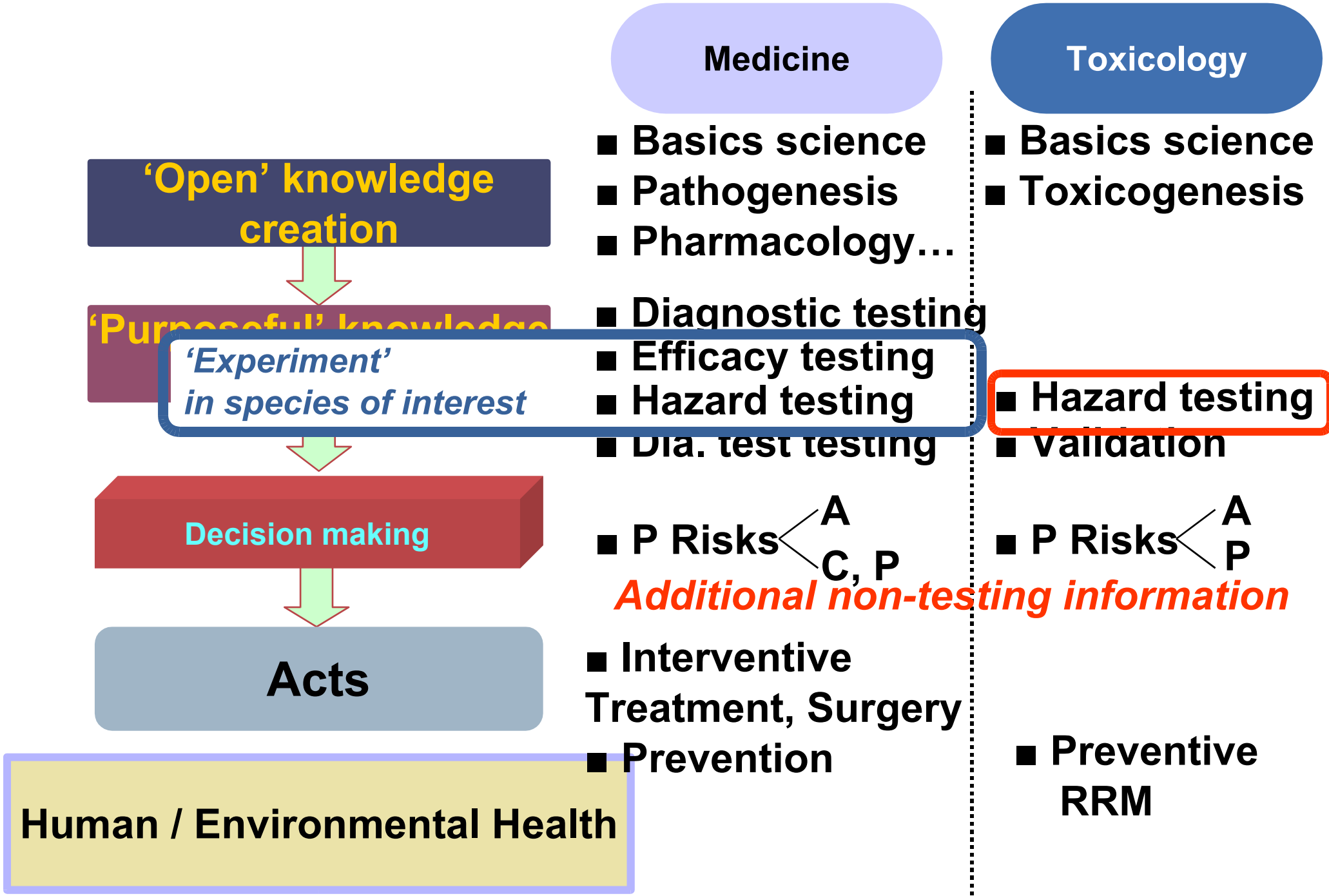
Curative/Preventive acts

Risk

Effectiveness

Core concepts: causation and probability

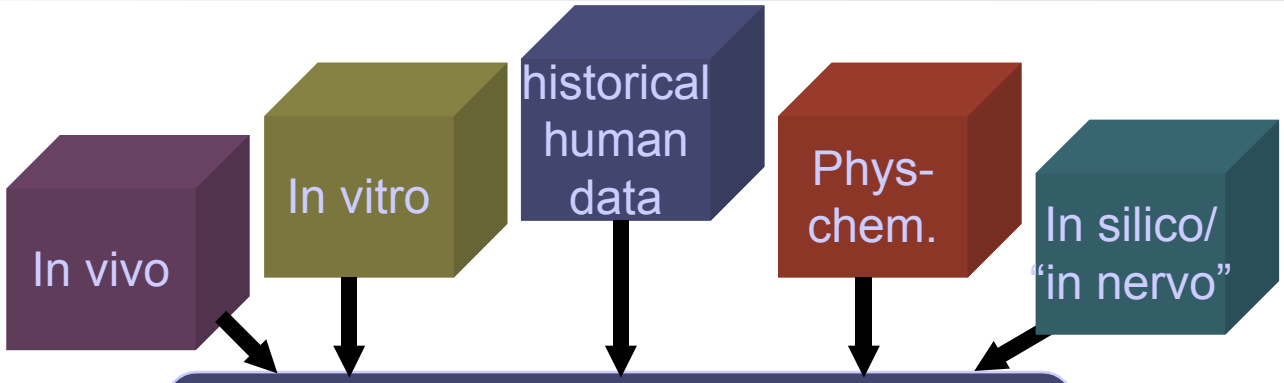
	Medicine	Toxicology
Causation	*Disease, hazard*	*Hazard*
	Pathogenesis	*"Toxicogenesis"*
	Curative/preventive acts	*Curative/Preventive acts*
Probability	*Risk (A, A2)*	*Risk (A)*
	Effectiveness	*Effectiveness*

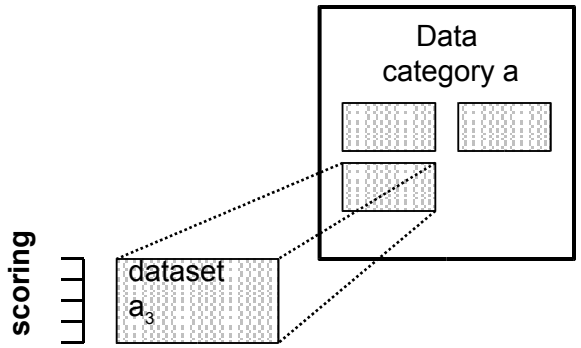

Medicine
Toxicology
‘Open’ knowledge creation
‘Purposeful’ knowledge
‘Experiment’
in species of interest
Decision making
Acts
Human / Environmental Health
■ Basics science
■ Pathogenesis
■ Pharmacology…
■ Diagnostic testing
■ Efficacy testing
■ Hazard testing
■ Dia. test testing
■ P Risks A C, P
■ Interventive Treatment, Surgery
■ Prevention
■ Basics science
■ Toxicogenesis
■ Hazard testing
■ Validation
■ P Risks A P
Additional non-testing information
■ Preventive RRM

Application in toxicology ?

- Developing and applying ex ante criteria for extracting "evidential power" of information to evaluate a specific question using structured approaches (systematic reviews)
- Quantitative data evaluation for acute decision making

Quantitative assessment of data (REACH)





Data evaluation, integration WoE judgment

Sufficient for classification ?

\+

Hazard (C&L)

Quantitative information

RISK

\-

In vitro

In vivo

Results

Declaration

10 defining characteristics

Definition / mission statement

Proceedings (early 2008)

EBT symposium Eurotox, Rhodes, Greece, 2008

Facilitation steps

Dissemination

Setting up method groups

Cross-fertilization with other e.b. disciplines

OVERVIEW

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CORRELATE…

- Is functional.
- Products will be peer-reviewed & endorsed by ESAC.
- 1st validation study starting December / January.
- Strong motivation to cooperate with national reference laboratories (to be set up).

Possible roles of CORRELATE

Validation tool for similar methods

Market 'pluralism' of methods

Reference laboratory network

European added value

Prospective validation

Assessment: robustness, handling qualities

REACH suitability

Continuous REACH implementation - ECHA

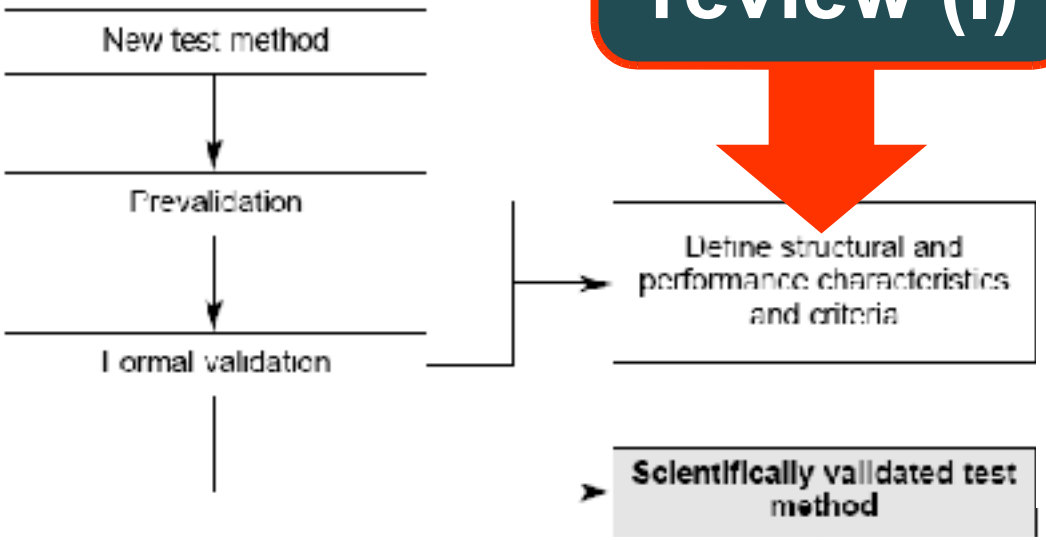
Two catch-up studies so far

Skin corrosion

3. EpiDerm (catch-up with EPISKIN)	2000
4. SkinEthic	2006
5. EST-1000	2007/8

The "classical" catch-up validation pathway

Figure 2: A schematic representat process



Peer review (I)

Peer review (II)

In the ECVAM process, a scientifically validated method is one that has been endorsed by the ECVAM Scientific Advisory Committee (ESAC). If the method is appropriate for chemicals testing, a draft Annex V guideline, incorporating the method, will be submitted to the EU Competent Authorities for Directive 67/548/EEC *for consideration for regulatory acceptance and application.*

Worth AP & Balls M

Catch-up vs me-too: between laboratory variability

Hypothesis 1: Similarity ✓

	Submitter	Correlate	Third laboratory	BLV datasets (required = 3)
SCENARIO 1	● ● ●			3
SCENARIO 2	● ●	●		3
SCENARIO 3	●	●	●	3

"Catch-up" process → ***ESAC peer review panel***

"Me too" process → **Peer review** → ***ESAC peer review panel***

ESAC peer review panel → **Validation statement**

Catch-up vs me-too: between laboratory variability

"Catch-up" process

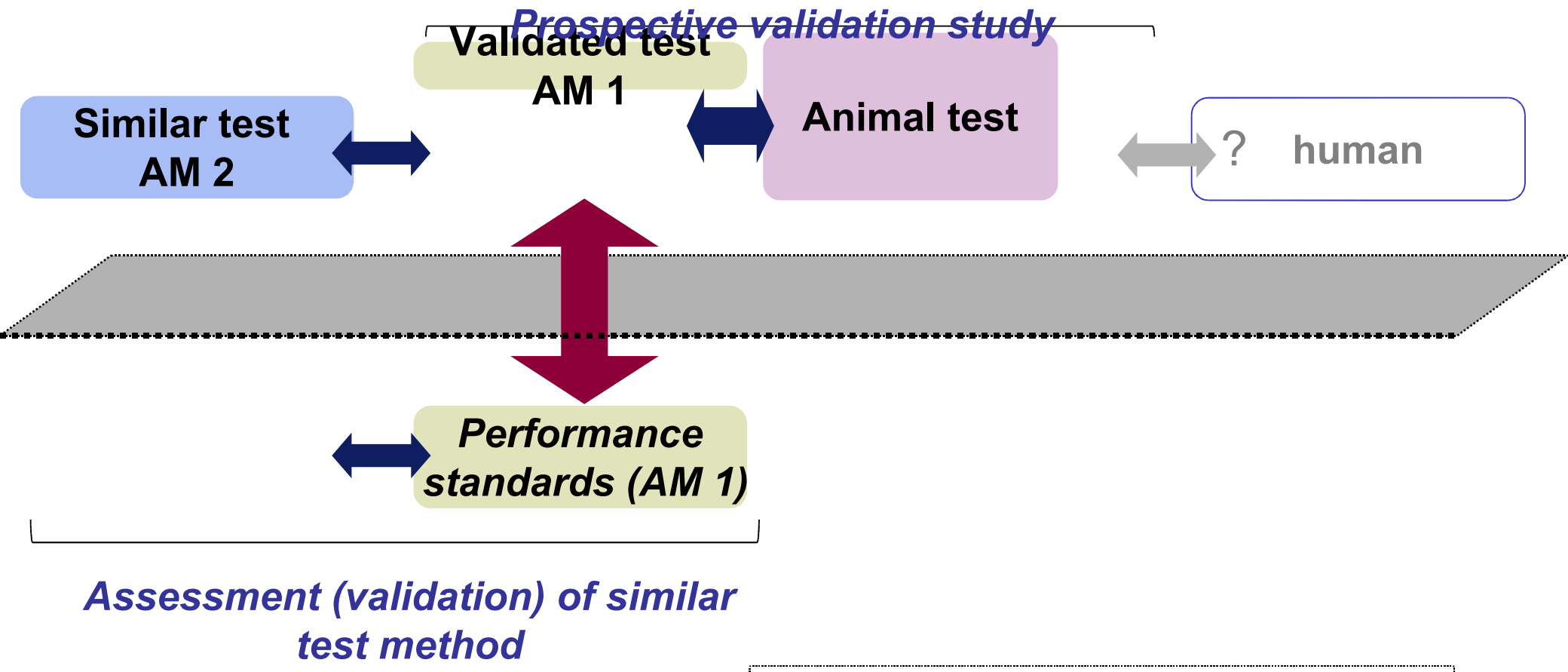
In the past:
Comparable to submissions of external validation studies. No independent laboratory involved.

In the future:
External studies coordinated by ECVAM with / without participation of NETWORK laboratory.

"Me too" process

In the future:
Studies with part of data generation by ECVAM-CORRELATE (and NETWORK laboratory)

The "validation regress" of justifications



Validation of similar methods

Performance standards

Hypothesis 1: Similarity

Conditional hypothesis !

Limited experimental set

Applicability domain

Hypothesis 2: Equal performance

Reproducibility

Predictive relevance

Validation of similar methods

Hypothesis 1: Similarity

Conditional hypothesis !

Limited experimental set

Hypothesis 2: Equal performance

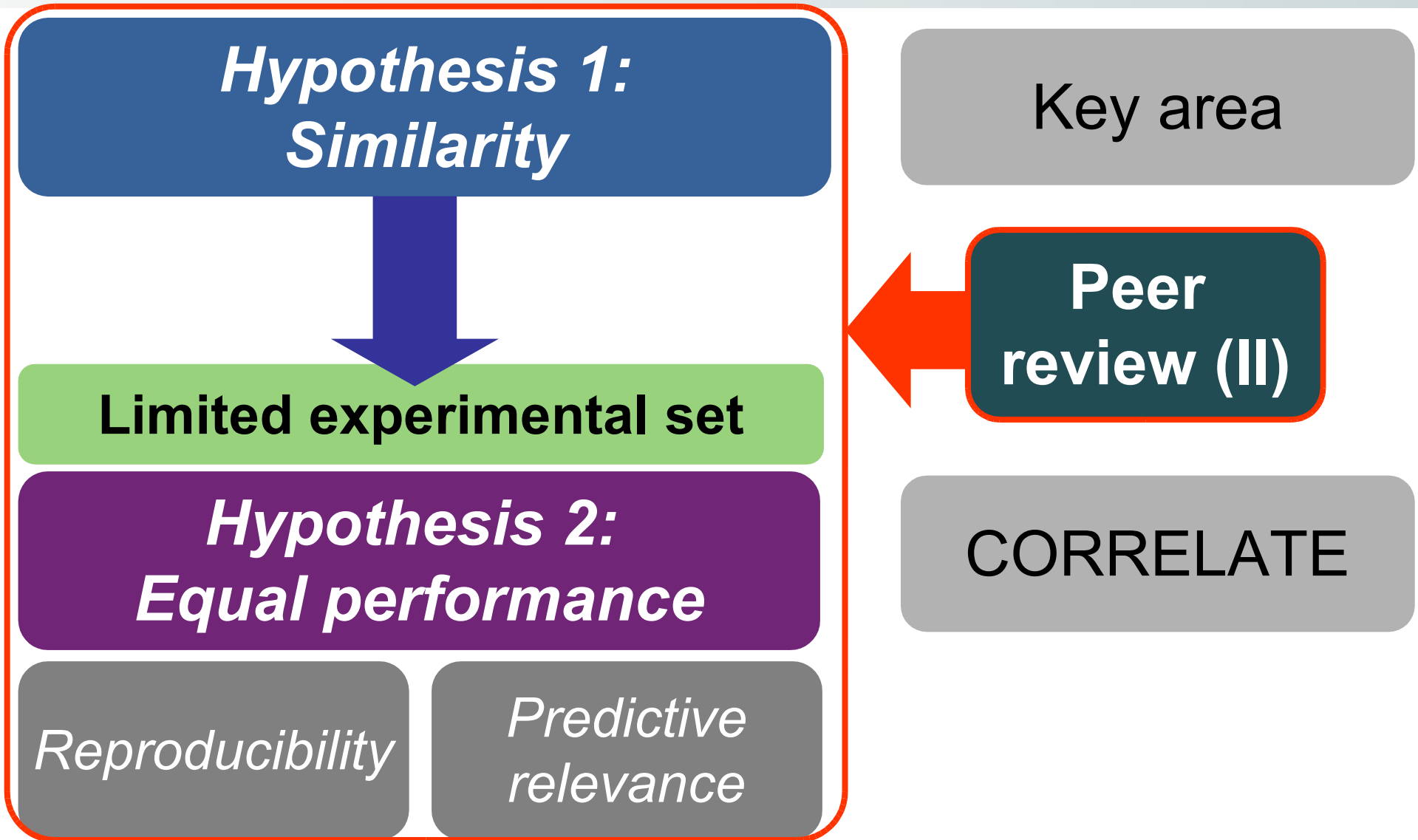
Reproducibility

Predictive relevance

NOT empirically testable. Justification via standards / ex ante criteria linked: Evidence-based

Empirically testable using a limited experimental set (reference chemicals)

Peer review is essential in evidence-based approaches



ESAC statements for new methods might be *updated* once similar methods have been validated

ESAC statement on scientific validity of

- **New method AM1**
- **Similar method AM2**
- **Similar method AM3**
- **Similar method AM4**

- ***version 1***
- ***version 2***
- ***version 3***
- ***version 4***

Summary (1)

Similar method

catch-up track

Me-too track (CORRELATE)

Standards

Applying these standards

Similarity: non empirical

Equal performance: empirical

ESAC endorsement in form of updating original statements

Peer review ✓

Peer review

Thank you for your attention !