PETA INTERNATIONAL SCIENCE CONSORTIUM LTD.

CW+ ChemicalRiskManager

Skin Irritation & Corrosion

25 January 2018

Webinar aims

- The first in a series of three webinars with the PETA International Science Consortium. This series updates the popular 2014-2015 webinar series and will reflect the significant progress in the use and acceptance of non-animal testing that has occurred in recent years. It will include:

- *In vitro* methods that can be used to meet REACH data requirements for skin irritation and corrosion under Annexes VII and VIII;

- Specific examples to explain how the in vitro methods can be used in an integrated approach to testing and assessment using specific examples.

Speakers

- **Dr Gertrude-Emilia Costin -** Gertrude-Emilia is the manager of scientific services at the Institute for In Vitro Sciences, Inc (IIVS). In her role as study director, she is in charge of a wide variety of safety and efficacy commercial studies based on in vitro test systems. Her main area of expertise is the use of in vitro test methods for the dermal safety assessment of ingredients and final formulations manufactured by the personal-care or pharmaceutical industries as well as that of products to be registered under the US Federal Insecticide, Fungicide, and Rodenticide Act, which is regulated by the US Environmental Protection Agency.

- **Dr Costanza Rovida –** Costanza is a chemist working as a scientific officer at the Center for Alternatives to Animal Testing–Europe (CAAT-Europe) and a project manager for TEAM Mastery. Convinced that in vivo methods are not the right scientific answer to our toxicological questions, from 2005 to 2008, she worked at the European Centre for the Validation of Alternative Methods, where she gained experience in the application of alternative methods in the area of skin and respiratory sensitisation.

Chair – Name – Job Title - Organisation

Questions

- **Please submit questions during the webinar using your chat box.**

- Any unanswered questions can be raised in the Chemical Watch LinkedIn group following the webinar:

www.chemicalwatch.com/CRM-LI

Event Title 2017

13-14 November | Brussels, Belgium

#Hash tag | @chemicalwatch

Skin Irritation/Corrosion Webinar

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25 January 2018

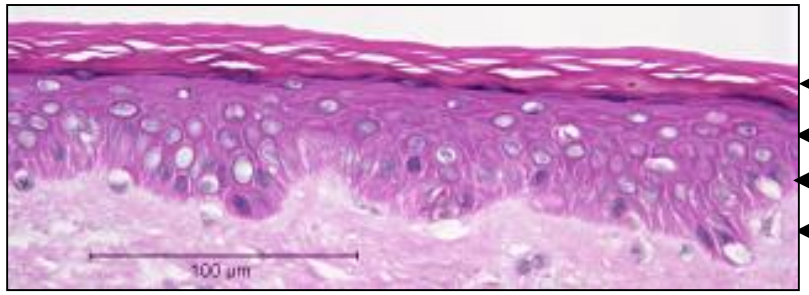
Overview

1. The impact of test substances on human skin
2. The animal test system used for skin corrosion and irritation assessment
3. Evaluation of skin corrosion and irritation potential using *in vitro* assays
4. Conclusions – *in vitro* assays validated for regulatory purposes (skin corrosion and irritation endpoints)

1. The impact of test substances on human skin

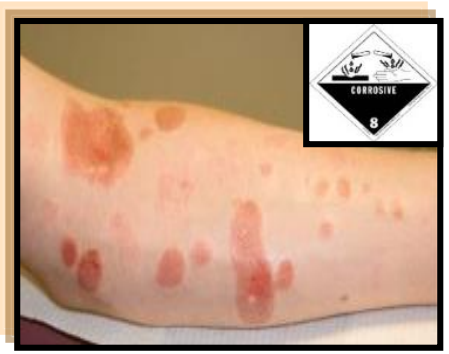
Skin corrosive/irritant

- Vast physical barrier against mechanical, chemical and microbial factors
- Immune network
- Unique defense system against UV irradiation



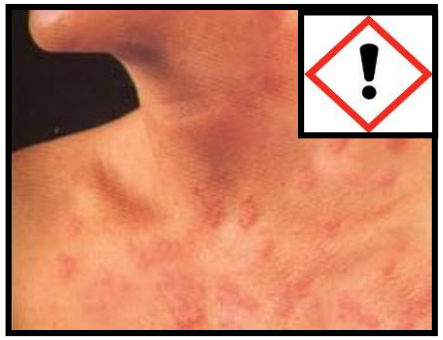
Stratum corneum
Stratum granulosum
Stratum spinosum
Basal layer of dividing keratinocytes

Native human skin



CORROSION

- Irreversible damage of the skin following exposure to a test substance
- Visible necrosis through the epidermis and into dermis - macroscopically typified by ulcers, bleeding, etc.



IRRITATION

- Reversible damage of the skin following exposure to a test substance
- Characterized macroscopically by erythema (redness) and oedema
- Damage to keratinocytes and dermal cells leads to inflammation

Registration and labeling of chemicals * Transport of chemicals * Occupational safety

Safety of cosmetics, toiletries and household products

http://www.survivingdisasters.info/emergency-first-aid/c/chemical-burn

2. The animal test system used for skin corrosion and irritation assessment

Acute dermal corrosion and irritation animal test (Draize)

Brief overview and current regulatory status

- **Test system**: albino rabbits
- **Assay endpoint**: erythema and eschar formation; oedema formation
- **Assay control**: untreated skin areas of the test animal

- **Applicability**: Evaluation of the corrosion and irritation potential of test substances
- **Limitations**: The rabbit and human skin have different physiological properties and responses to environmental and chemical agents. Some test substances are more toxic to rabbits than to humans and vice versa.

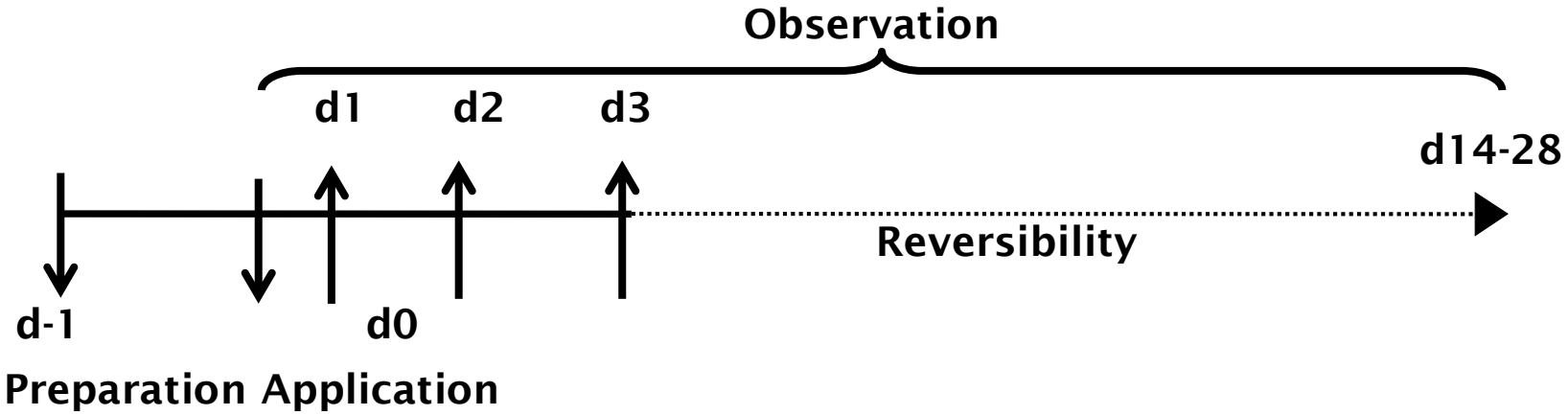
  The Draize rabbit skin irritation test has been criticized for over-prediction of human skin irritation.

  A debated ethical issue of the *in vivo* test concerns the animals' suffering and discomfort.

- **Regulatory status**: OECD Test Guideline 404 (TG 404) (updated 24 April 2015)

Draize: Typical protocol

- **Animals:** 1-3 rabbits (sequential testing)
- **Test substance:** 0.5 mL or 0.5 g of usually undiluted liquid or moistened solid test substance applied on 6 cm² skin surface
- **Exposure**: 3 minutes, 1 hr or 4 hr (skin corrosion);
  4 hr (skin irritation)



Erythema and eschar formation	
No erythema	0
Very slight erythema (barely perceptible)	1
Well defined erythema	2
Moderate to severe erythema	3
Severe erythema (beef redness) to eschar formation preventing grading of erythema	4

Oedema formation	
No oedema	0
Very slight oedema (barely perceptible)	1
Slight oedema (edges or area well defined by definite raising)	2
Moderate oedema (raised approximately 1 mm)	3
Severe oedema (raised more than 1 mm and extending beyond area of exposure	4

Draize J. H., Woodard G., Clavery, H.O. Methods for the study of irritation and toxicity of substances applied topically to the skin and mucous membranes, J. Pharmacol. Exp. Ther., ***82****, 377-390 (1944).*

3. Evaluation of skin corrosion and irritation potential using *in vitro* assays

❖ *General considerations*

Assessment of skin corrosion and irritation

- The impact of test substances on skin is evaluated progressively from corrosion and irritation potential to human compatibility
- Various assays are available depending on the purpose of the testing

In vitro assays validated for regulatory purposes	OECD TG
SKIN CORROSION	
• Transcutaneous Electrical Resistance (TER) Test • RhE (Reconstructed human EpiDermis) Test Method • Membrane Barrier Test Method (Corrositex®)	430 431 435
SKIN IRRITATION	
• RhE Test Method – SIT (Skin Irritation Test)	439

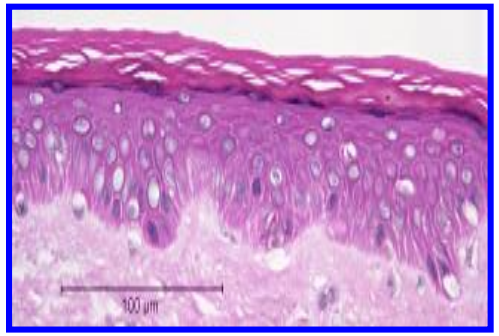
Hazard identification and labelling of chemicals
Transportation of dangerous goods like industrial chemicals (neat or diluted) and their mixtures
Labelling of finished products (cleaning agents, household products)

In vitro reconstructed human epidermis (RhE) models validated for regulatory purposes

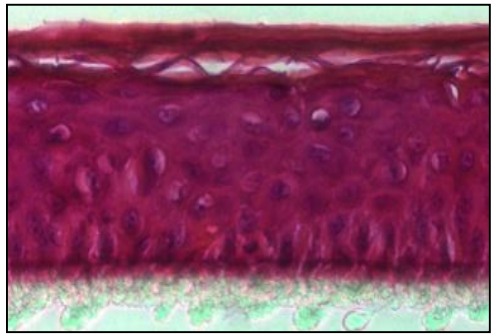
1. General model criteria

- Human keratinocytes are used for construction of the models
- Multiple layers of viable epithelial cells
- Functional *stratum corneum* and barrier

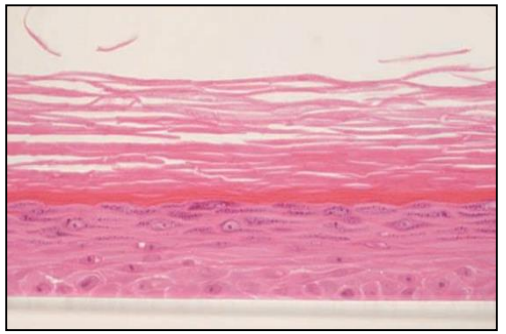
Native human skin



epiCS®



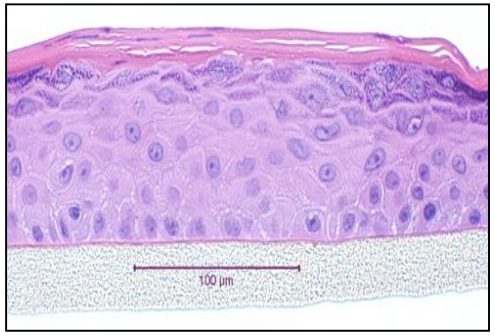
LabCyte EPI-MODEL



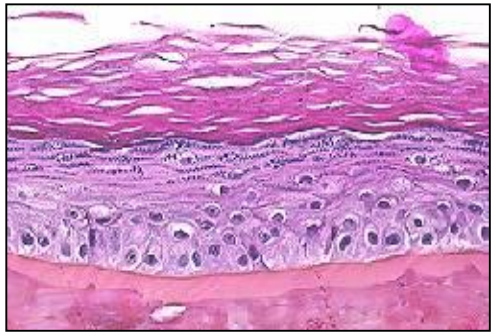
2. Functional model criteria

- Tissues must be viable (QC from manufacturer, internal controls)
- *Stratum corneum* must form sufficient barrier
- RhE models should exhibit long term reproducibility

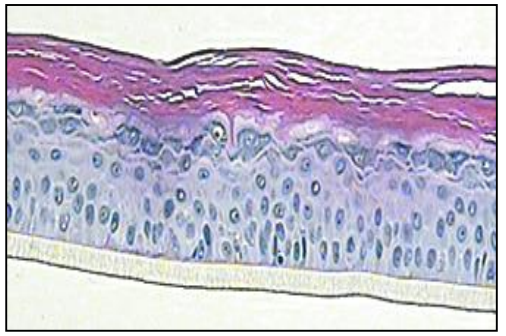
EpiDerm™ (EPI-200)



EpiSkin™ (SM)



SkinEthic™ RHE



3. Evaluation of skin corrosion and irritation potential using *in vitro* assays

❖ *Assay specific considerations*

***In vitro* skin corrosion**

Transcutaneous Electrical Resistance Test (OECD 430)

Brief overview and current regulatory status

- **Test system**: rat skin discs (dorso-lateral, 20 mm each, tested in triplicate from the same animal) - from humanely killed rats aged 28-30 days; Wistar-derived or comparable strain
- **Assay endpoint**: electrical impedance of the skin expressed as a transcutaneous electrical resistance (TER) value in kilo Ohms (kΩ) – measure of barrier function
- **Assay controls**: negative (sterile, deionized water); positive (10 M hydrochloric acid)

- **Applicability**: Identification of non-corrosive and corrosive test substances and mixtures in accordance with the UN GHS (Globally Harmonized System)
- **Limitations**: Does not allow the sub-categorization of corrosive substances and mixtures in accordance with the UN GHS

- **Regulatory status**: OECD Test Guideline 430 (TG 430) (updated 26 July 2015)

TER: Typical protocol

Tissue Preparation

Skin discs of ~20 mm each are prepared from rats using the procedure outlined in the OECD TG 430.

Tissue Mounting

Each skin disc is placed over one end of a PTFE (polytetrafluoroethylene) tube, with the epidermal surface in contact with the tube.

Tissue Treatment

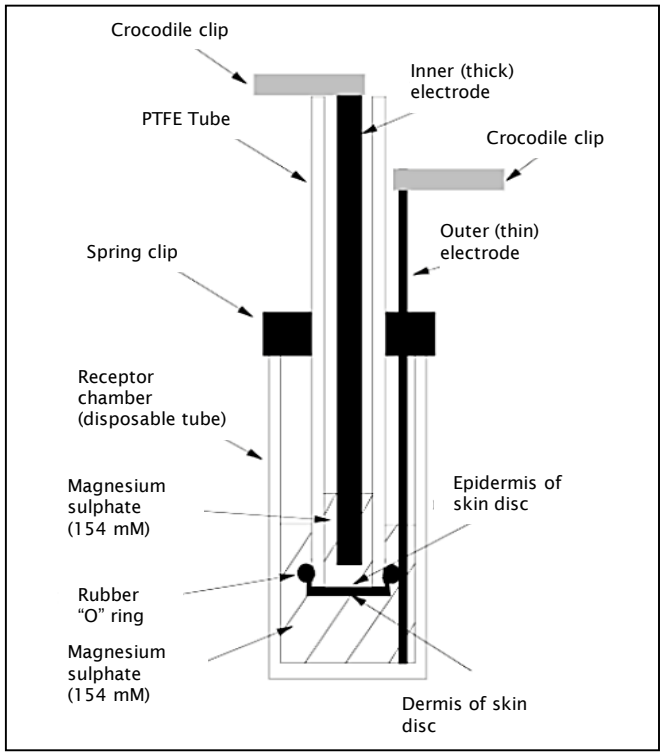
Test substances are applied to the epidermal surface inside the tube for 24 hr at 20-23°C (150 µL for liquids test substances; 150 µL di water added on top of solid test substances).

Treatment Termination

The test substance is removed with a jet of tap water at up to 30°C.

TER Measurement

The skin impedance is measured as TER by using a low-voltage, alternating current Wheatstone bridge (1-3V operating voltage, sinus or rectangular shaped AC of 50-100 Hz, measuring range of at least 0.1-30 kΩ).



Prediction Model

Resistance measured	Prediction to be considered – UN GHS Category
i) If the mean TER value obtained for the test substance is ≤ 5 kΩ and the skin discs are obviously damaged (*e.g.*, perforated), **OR** ii) The mean TER value obtained for the test chemical is ≤ 5 kΩ, **AND** • The skin discs show no obvious damage (*e.g.*, perforation), **BUT** • The mean discs dye content is ≥ the mean disc dye content of the 10M HCl positive control obtained concurrently)	Corrosive
i) If the mean TER value obtained for the test substance is > 5 kΩ, **OR** ii) The mean TER value obtained for the test substance is ≤ 5 kΩ, **AND** • The skin discs show no obvious damage (*e.g.*, perforation), **AND** • Thea mean disc dye content is < the mean disc dye content of the 10 M HCl positive control obtained concurrently	Non-corrosive

Dye Binding Method

For test substances that can remove the skin lipids (detergents, emulsifiers, etc.) and make the skin barrier more permeable to ions: if TER values are ~5 kΩ, an assessment using 150 µL of the dye sulphorhodamine B (10% w/v) should be performed (2 hr). If the stratum corneum is compromised, the dye will rapidly penetrate and the amount of the dye bound to the skin is assessed (µg/disc) by reading the Optical Density (OD_{565nm}).

RhE Test Method - Skin Corrosion Assay (OECD TG 431)

Brief overview and current regulatory status

- **Test system:** RhE models [EpiDerm™ (EPI-200); EpiSkin™ (SM); SkinEthic™, RHE; and epiCS®]
- **Assay endpoint:** tissue viability (%) – assessed by reduction of the vital dye MTT (3-[4,5-dimethylthiazol-2-yl]-2,5-diphenyltetrazolium bromide) by viable cells
- **Assay controls:** negative (sterile, deionized water or NaCl solution 9g/L)
  positive (8N KOH or glacial acetic acid – only for 4 hr exposure)

- **Applicability:** The results can be used for regulatory purposes for distinguishing or sub-categorization, *i.e.*, 1A vs. 1B-and-1C vs. non-corrosive test substances.
- **Limitations:** The method does not allow discriminating between skin corrosive sub-categories 1B and 1C according to the UN GHS due to a limited set of well-known *in vivo* corrosive Category 1C chemicals.

- **Regulatory status:** OECD Test Guideline 431 (TG 431) (updated 29 July 2016)

RhE: Typical protocol

Tissue Receipt

Upon receipt, tissues are incubated for at least 1 hr in standard culture conditions (37±1°C in a humidified atmosphere of 5±1% CO_2 in air).

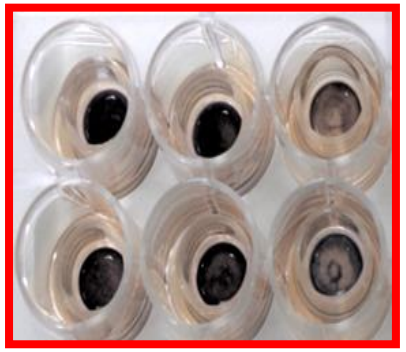
Tissue Treatment

Media is refreshed after the initial 1 hr incubation. Duplicate tissues are treated topically with control and test substances for 3 min / 1 hr (4 hr) (50 µL dose).

Tissue Rinsing

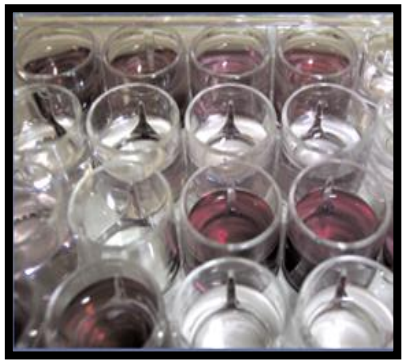
After exposure, tissues are rinsed to remove the control and test substances.

MTT Reduction



Individual tissues are placed into wells containing unreduced MTT solution and incubated at standard culture conditions for 3 hr.

Isopropanol Extraction



The tissues are placed in isopropanol at room temperature for 2 hr to extract the reduced MTT. Extracted MTT is thoroughly mixed and transferred to a 96-well plate.

Spectrophotometric Quantification

Optical density (OD) at 570 nm (OD_{570}) is determined using a 96-well plate reader. OD values are used to calculate relative viability values presented relative to negative control tissue values.

RhE-Corrosion: Prediction Models

EpiSkin™ (SM)

Viability measured after exposure time points (3, 60 and 240 minutes)	Prediction to be considered UN GHS Category
< 35% after 3-minutes exposure	Corrosive: • Optional Sub-category 1A
≥ 35% after 3-minutes exposure **AND** < 35% after 60-minutes exposure **OR** ≥ 35% after 60-minutes exposure **AND** < 35% after 240-minutes exposure	Corrosive: • A combination of optional Sub-categories 1B and 1C
≥ 35% after 240-minutes exposure	Non-corrosive

EpiDerm™ (EPI-200)
SkinEthic™ RHE
epiCS®

Viability measured after exposure time points (3- and 60-minutes)	Prediction to be considered UN GHS Category
STEP 1	
< 50% after 3-minutes exposure	Corrosive
≥ 50% after 3-minutes exposure **AND** < 15% after 60-minutes exposure	Corrosive
≥ 50% after 3-minutes exposure **AND** ≥ 15% after 60-minutes exposure	Non-corrosive
STEP 2	
<25%; 18%; 15% after 3-minutes exposure	Optional Sub-category 1A
≥25%; 18%; 15% after 3-minutes exposure	A combination of optional Sub-categories 1B-and-1C

Desprez B., Barroso J., Griesinger C., Kandarova H., Alepee N., Fuchs H.W., Two novel prediction models improve prediction of skin corrosive sub-categories by test methods of OECD, Toxicology in Vitro, 29, 2055-2080 (2015) .

Membrane Barrier Test Method (Corrositex®) (OECD 435)

Brief overview and current regulatory status

- **Test system**: artificial membrane designed to respond to corrosive substances in a manner similar to animal skin *in situ*
- **Assay endpoint**: the time (in minutes) required for a test substance to penetrate through the Corrositex Biobarrier Membrane and produce a color change in the Chemical Detection System (CDS)
- **Assay controls**: negative (10% citric acid, 5% propionic acid); positive (sodium hydroxide)

- **Applicability**: Assigns UN Packing Group to corrosives or verifies if a test substance is non-corrosive
- **Limitations**: Materials with a pH of ≥ 4.5 and ≤ 8.5 generally fail to qualify for testing based on the CDS used in the kit provided by In Vitro International

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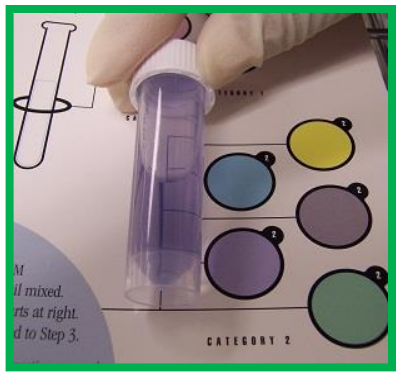
- **Regulatory status**: OECD Test Guideline 435 (TG 435) (updated 19 July 2015)

Corrositex®: Typical protocol

Qualification of the Test Substance

Each test substance is added to a vial containing the Chemical Detection System (CDS) to determine if the test substance will qualify for testing. A color change can be detected when the pH of the CDS drops below 4.5 or rises above 8.5.

Categorization



The test substance is next categorized to determine the appropriate timetable for Packing Group Assignment. The test substance is added to two tubes. The resulting color change in each tube is compared to the color chart provided by In Vitro International. A category 1 test substance will be evaluated for up to 4 hr; a category 2 test substance will be evaluated for up to 1 hr.

Biobarrier Preparation

To prepare the biobarrier membranes, the biobarrier matrix powder is completely solubilized. The solubilized collagen matrix, composed of water and dissolved proteins, is then added to a membrane disc containing a porous cell membrane. The biobarrier membrane is placed onto a vial of CDS.

Biobarrier Placement

Break Through Observations

Each test substance is added onto four replicate biobarrier membranes and the CDS vial is continuously monitored for the first 10 min. If no color change occurs, this process is repeated an additional three times until the remaining biobarrier membranes are treated with the test substance. The vials are observed until a color change (*i.e.*, break through) occurs. When a color change occurs in each vial, the break through times are recorded.

Corrositex®: Prediction Model

Category I

Mean Time to Produce a Change in Chemical Detection System	Packing Group
≤ 3 Minutes	I
> 3 Minutes - 1 Hour	II
> 1 - 4 Hours	III
> 4 Hours	Not Applicable

Category II

Mean Time to Produce a Change in Chemical Detection System	Packing Group
≤ 3 Minutes	I
> 3 Minutes - 30 minutes	II
> 30 - 60 minutes	III
> 60 minutes	Not Applicable

3. Evaluation of skin corrosion and irritation potential using *in vitro* assays

❖ *Assay specific considerations*

***In vitro* skin irritation**

RhE Test Method (SIT) (OECD TG 439)

Brief overview and current regulatory status

- **Test system**: RhE models (EpiDerm™ (EPI-200), EpiSkin™ (SM), SkinEthic™ RHE, LabCyte EPI-MODEL24)
- **Assay endpoint**: tissue viability (%) – MTT
- **Assay controls**: negative (sterile, deionized water or Calcium and Magnesium Free DPBS); positive (5% SDS)

- **Applicability**: The results can be used for regulatory purposes to determine the skin irritancy of test substances either as a stand-alone replacement for *in vivo* skin irritation testing or as partial replacement test within a tiered testing strategy
- **Limitation**: Does not allow the classification of test substances to the optional UN GHS Category 3 (mild irritants).

- **Regulatory status**: OECD Test Guideline 439 (TG 439) (updated 26 July 2015)

RhE: Typical protocol

Tissue Receipt

Upon receipt, tissues are incubated first for 1 hr and then over night (with media change) in standard culture conditions.

Tissue Treatment

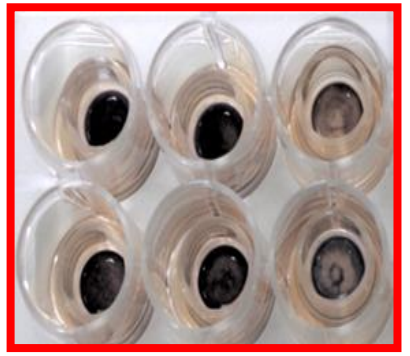
Triplicate tissues are treated topically with control and test substances (30 µL dose for liquids; 25 mg for solids).

Tissue Rinsing

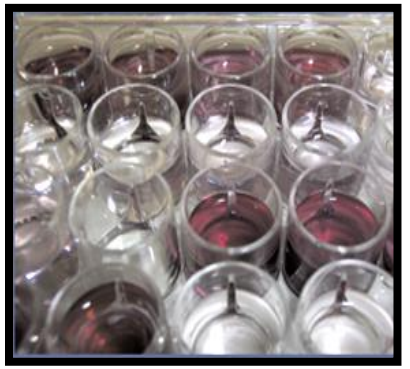
After exposure, tissues are rinsed and then placed in the incubator at standard culture conditions for an initial post-treatment incubation of 24±1 hr. After the initial post-treatment expression incubation, the tissues are transferred in fresh medium and placed back in the incubator for the remainder of the 42±2 hr post-treatment incubation (to capture delayed effects of test substances on the reconstructed tissues).

Post-treatment Expression Incubation

MTT Reduction



Isopropanol Extraction



Spectrophotometric Quantification

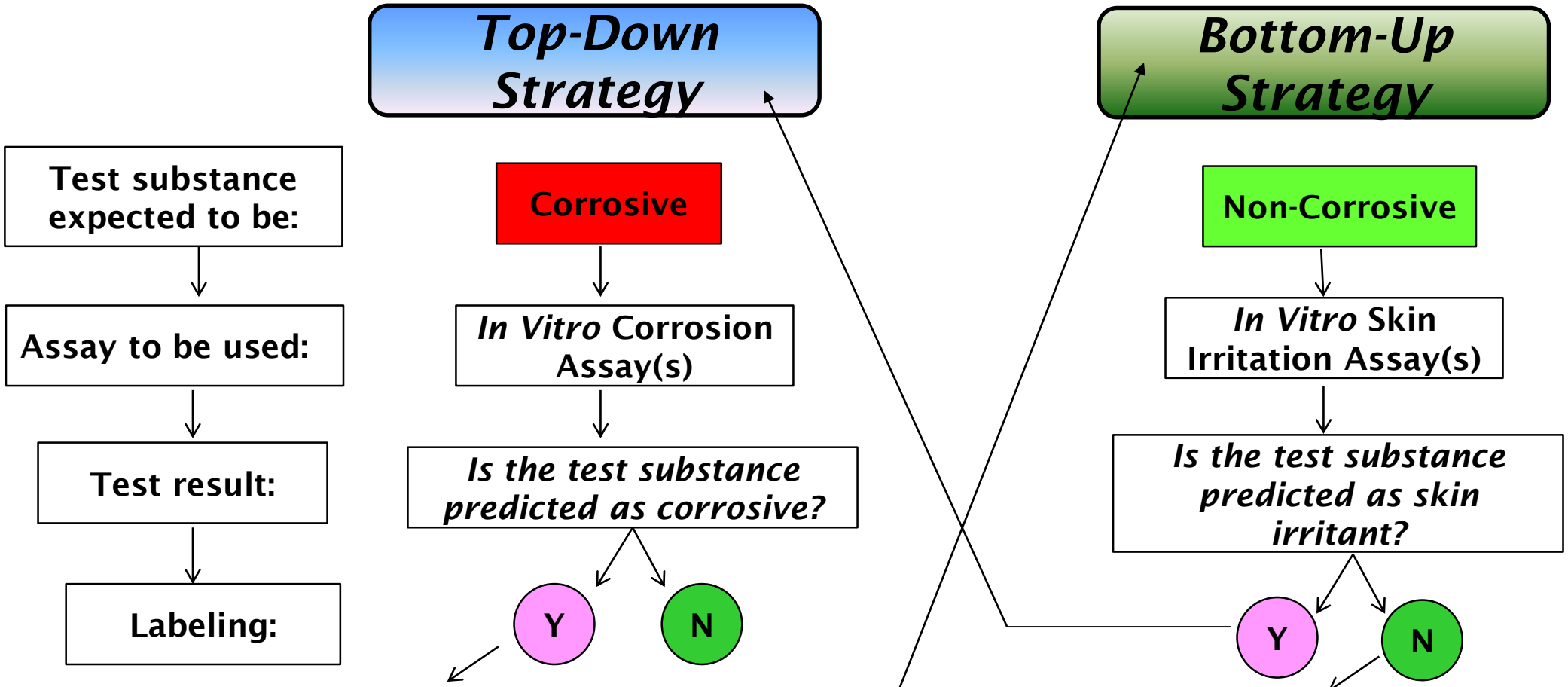
Prediction Model

In vitro result	*In vivo* prediction	UN GHS CATEGORY
Mean tissue viability ≤ 50%	Irritant (I)	**Category 1 or 2** (*OECD 431 to be used for confirmation of Category 1*)
Mean tissue viability > 50%	Non-irritant (NI)	***No Category***

4. Conclusions – *in vitro* assays validated for regulatory purposes (skin corrosion and irritation endpoints)

- Assays designed for regulatory purposes often rely on a single exposure time / dose which provides a predictive response
- Assays designed for regulatory use often are limited to the current *in vivo* assay range
- Frequently ingredients-oriented
- Limited in their predictive scope (*not useful for evaluating toxic effects outside of specific predictive range*)
- In the interest of both sound science and animal welfare, *in vivo* testing should not be undertaken until all available data relevant to the potential dermal corrosion and irritation of the test substance have been evaluated in a weight-of-the-evidence analysis.
- Tiered testing strategy including:
  - search for existing studies in humans and/or laboratory animals
  - evidence of corrosion and irritation of structurally related substances or mixtures
  - data demonstrating strong acidity or alkalinity of the substance
  - results from validated and accepted *in vitro* or *ex vivo* tests

Tiered testing strategies for the assessment of skin corrosion and irritation potential



- Test substance may be labeled as corrosive.
- Test substance may also be classified by the relevant packing group.

The test substance does not require any further skin irritation testing for submission to regulatory agencies utilizing the GHS system.

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CAAT eu
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Johns Hopkins Bloomberg School of Public Health

IIVS Institute for In Vitro Sciences
PETA International Science Consortium Ltd.

SKIN IRRITATION AND CORROSION

25th January 2018

#Hash tag | @chemicalwatch

SKIN IRRITATION AND CORROSION

Testing in vitro for regulatory purposes

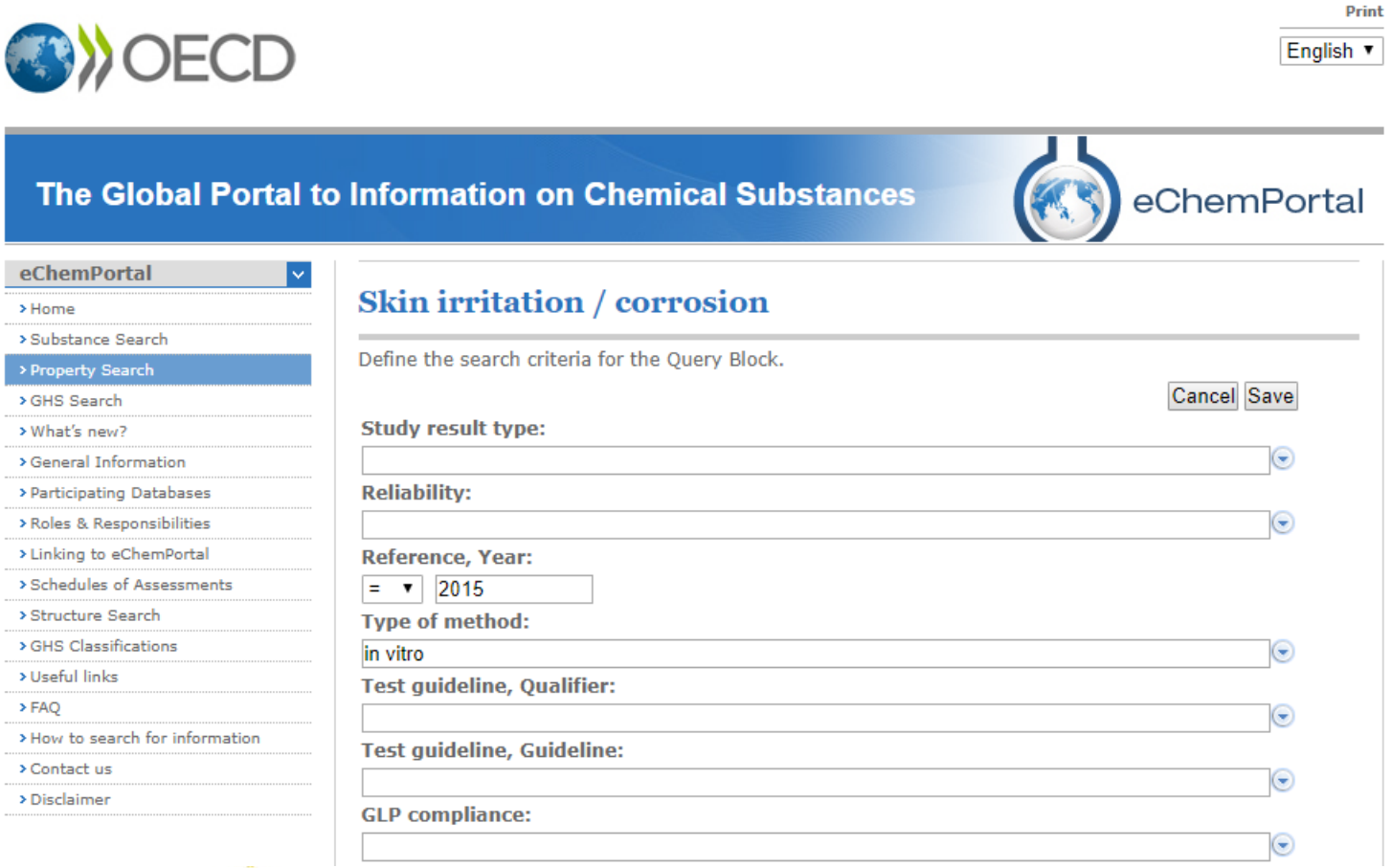
Costanza Rovida

CAAT Europe

TEAM Mastery

costanza.rovida@chimici.it

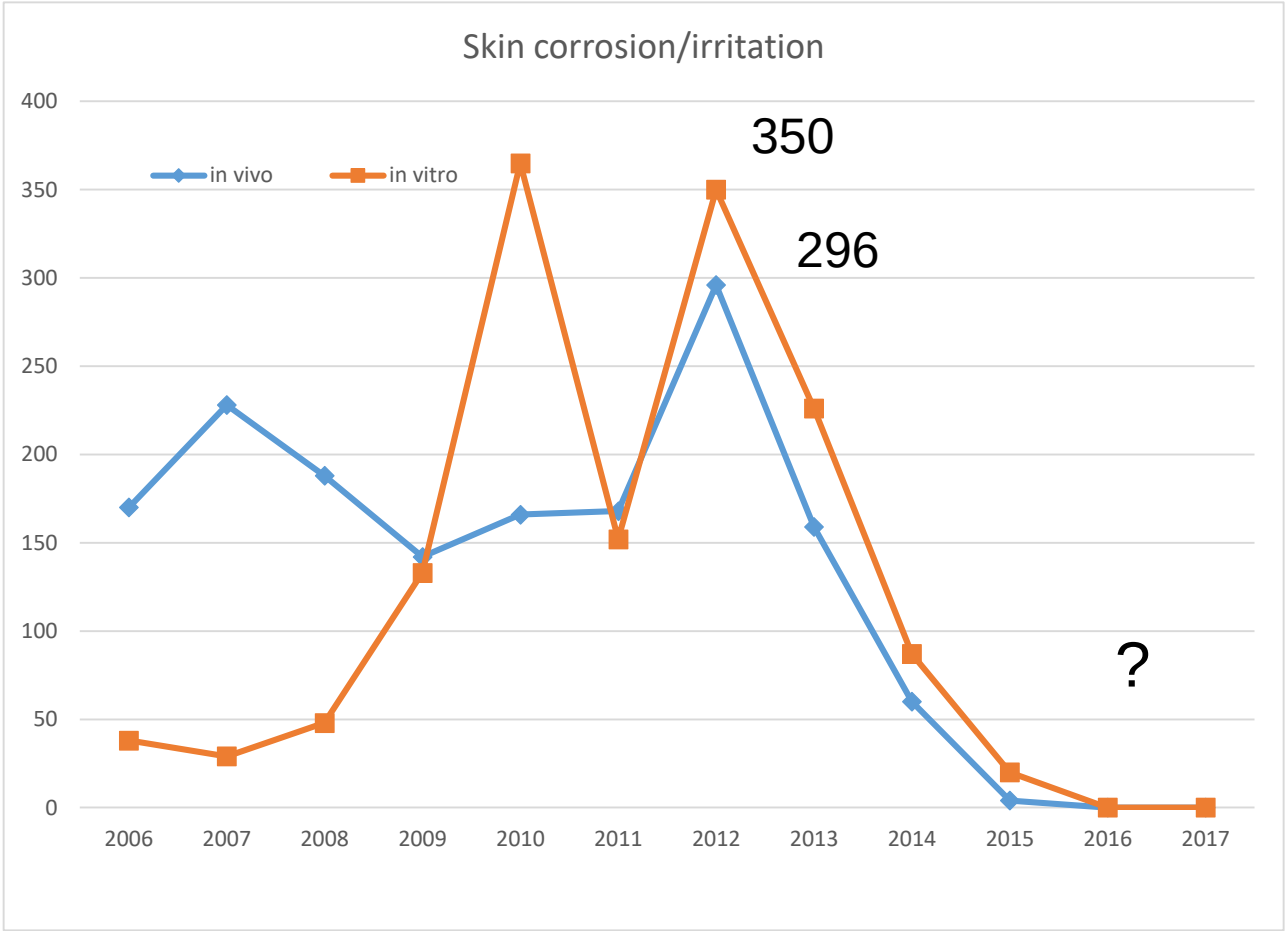
www.echemportal.org



Number of tests per year

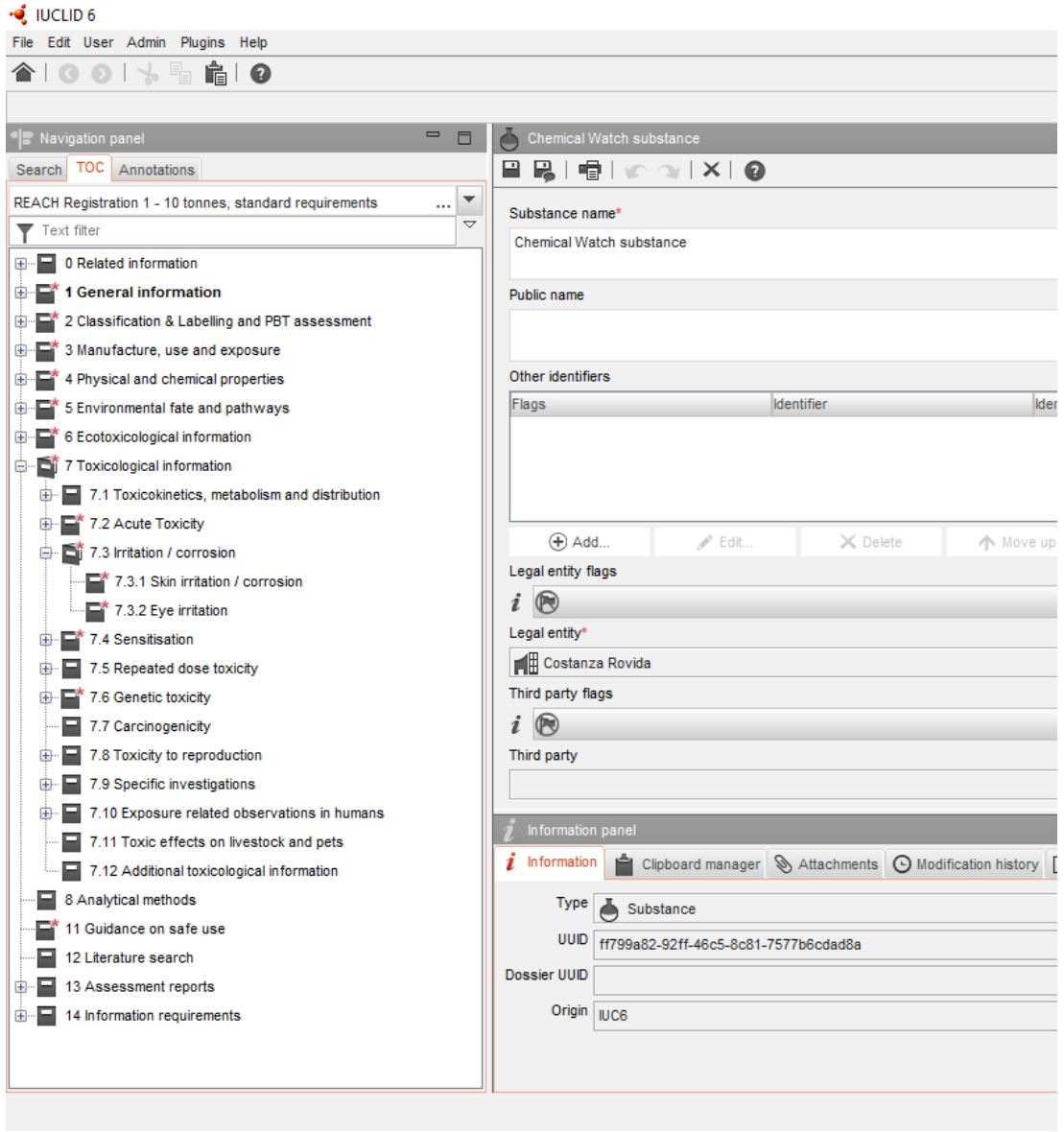
REACH Registration deadlines

- 30th November 2010
  substances > 1000 t/y
  CMR > 1 t/y
- 31st May 2013
  substances > 100 t/y
- 31st May 2018
  substances > 1 t/y



REACH Regulation, EC 1907/2006
(Registration, Evaluation, Authorisation and restriction of Chemicals)

https://iuclid6.echa.europa.eu/



TEAM mastery
Know the rules, play your market.

CAAT eu

REACH and the use of alternative methods

Article 1:

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

Article 13:

In particular for human toxicity, information shall be generated whenever possible by means other than vertebrate animal tests, through the use of alternative methods, for example, in vitro methods or qualitative or quantitative structure-activity relationship models or from information from structurally related substances (grouping or read-across).

Annex VI "STEP 4 — GENERATE NEW DATA/PROPOSE TESTING STRATEGY

In some cases it will not be necessary to generate new data. However, where there is an information gap that needs to be filled, new data shall be generated (Annexes VII and VIII), or a testing strategy shall be proposed (Annexes IX and X), depending on the tonnage. New tests on vertebrates shall only be conducted or proposed as a last resort when all other data sources have been exhausted.

ANNEX XI

GENERAL RULES FOR ADAPTATION OF THE STANDARD TESTING REGIME SET OUT IN ANNEXES VII TO X

1.4. In vitro methods

…….

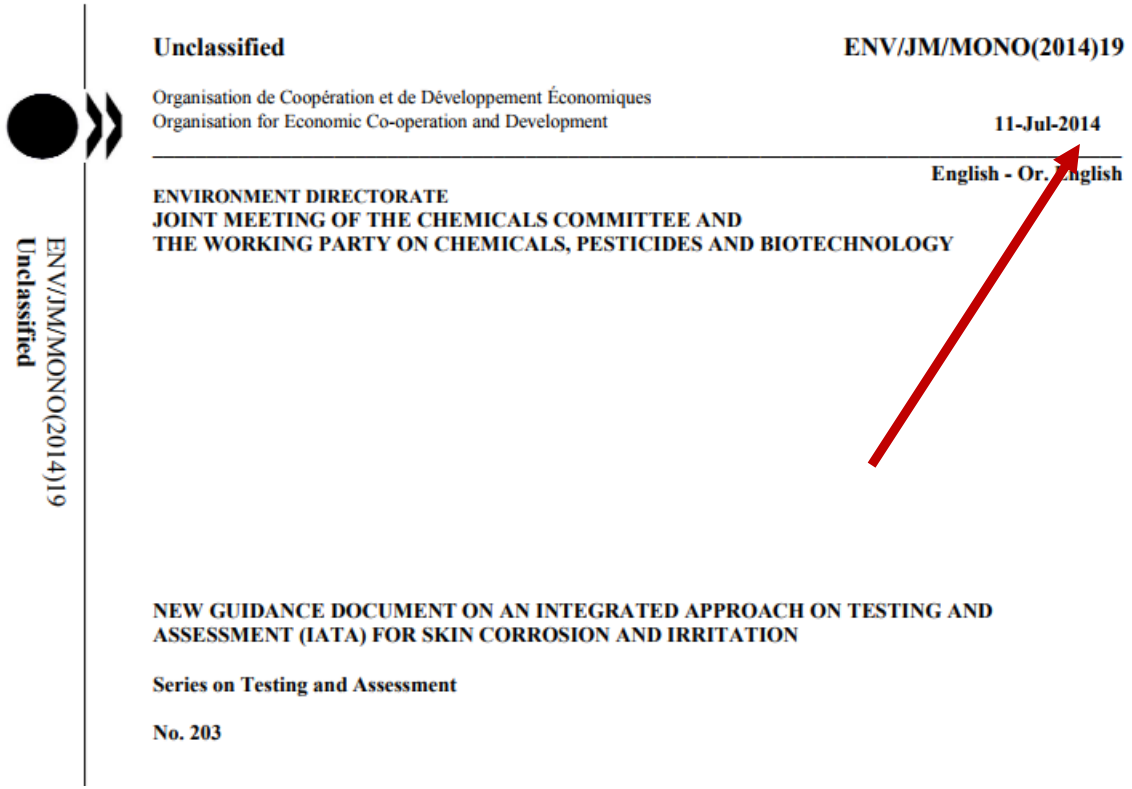
If the results obtained from the use of such in vitro methods do not indicate a certain dangerous property, the relevant test shall nevertheless be carried out at the appropriate tonnage level to confirm the negative result, unless testing is not required in accordance with Annexes VII to X or the other rules in this Annex. **Such confirmation may be waived**, if the following conditions are met:

(1) results are derived from an in vitro method whose scientific validity has been established by a validation study, according to internationally agreed validation principles;

(2) results are adequate for the purpose of classification and labelling and/or risk assessment; and

(3) adequate and reliable documentation of the applied method is provided.

OECD Guidelines



Unclassified ENV/JM/MONO(2014)19

Organisation de Coopération et de Développement Économiques
Organisation for Economic Co-operation and Development

11-Jul-2014

English - Or. English

ENVIRONMENT DIRECTORATE
JOINT MEETING OF THE CHEMICALS COMMITTEE AND
THE WORKING PARTY ON CHEMICALS, PESTICIDES AND BIOTECHNOLOGY

NEW GUIDANCE DOCUMENT ON AN INTEGRATED APPROACH ON TESTING AND ASSESSMENT (IATA) FOR SKIN CORROSION AND IRRITATION

Series on Testing and Assessment

No. 203

ENV/JM/MONO(2014)19
Unclassified

OECD TG 430 – first publication in 2004
***In Vitro* Skin Corrosion: Transcutaneous Electrical Resistance Test Method (TER)**

OECD TG 431 – first publication in 2004
***In Vitro* skin corrosion: reconstructed human epidermis (RHE) test method**

OECD TG 435 – first publication in 2006
***In Vitro* Membrane Barrier Test Method for Skin Corrosion**

OECD TG 439 – first publication in 2010
***In Vitro* Skin Irritation: Reconstructed Human *Epidermis* Test Method**

COMMISSION REGULATION (EU) 2016/863

of 31 May 2016

amending Annexes VII and VIII to Regulation (EC) No 1907/2006 of the European Parliament and of the Council on the Registration, Evaluation, Authorisation and Restriction of Chemicals (REACH) as regards skin corrosion/irritation, serious eye damage/eye irritation and acute toxicity

(Text with EEA relevance)

Annex VII (< 10 t/y)

'8.1. Skin corrosion/irritation	8.1. The study/ies do(es) not need to be conducted if: — the substance is a strong acid (pH ≤ 2,0) or base (pH ≥ 11,5) and the available information indicates that it should be classified as skin corrosion (Category 1), or — the substance is spontaneously flammable in air or in contact with water or moisture at room temperature, or — the substance is classified as acute toxicity by the dermal route (Category 1), or — an acute toxicity study by the dermal route does not indicate skin irritation up to the limit dose level (2 000 mg/kg body weight). If results from one of the two studies under point 8.1.1 or 8.1.2 already allow a conclusive decision on the classification of a substance or on the absence of skin irritation potential, the second study need not be conducted.
8.1.1. Skin corrosion, *in vitro*	
8.1.2. Skin irritation, *in vitro*	

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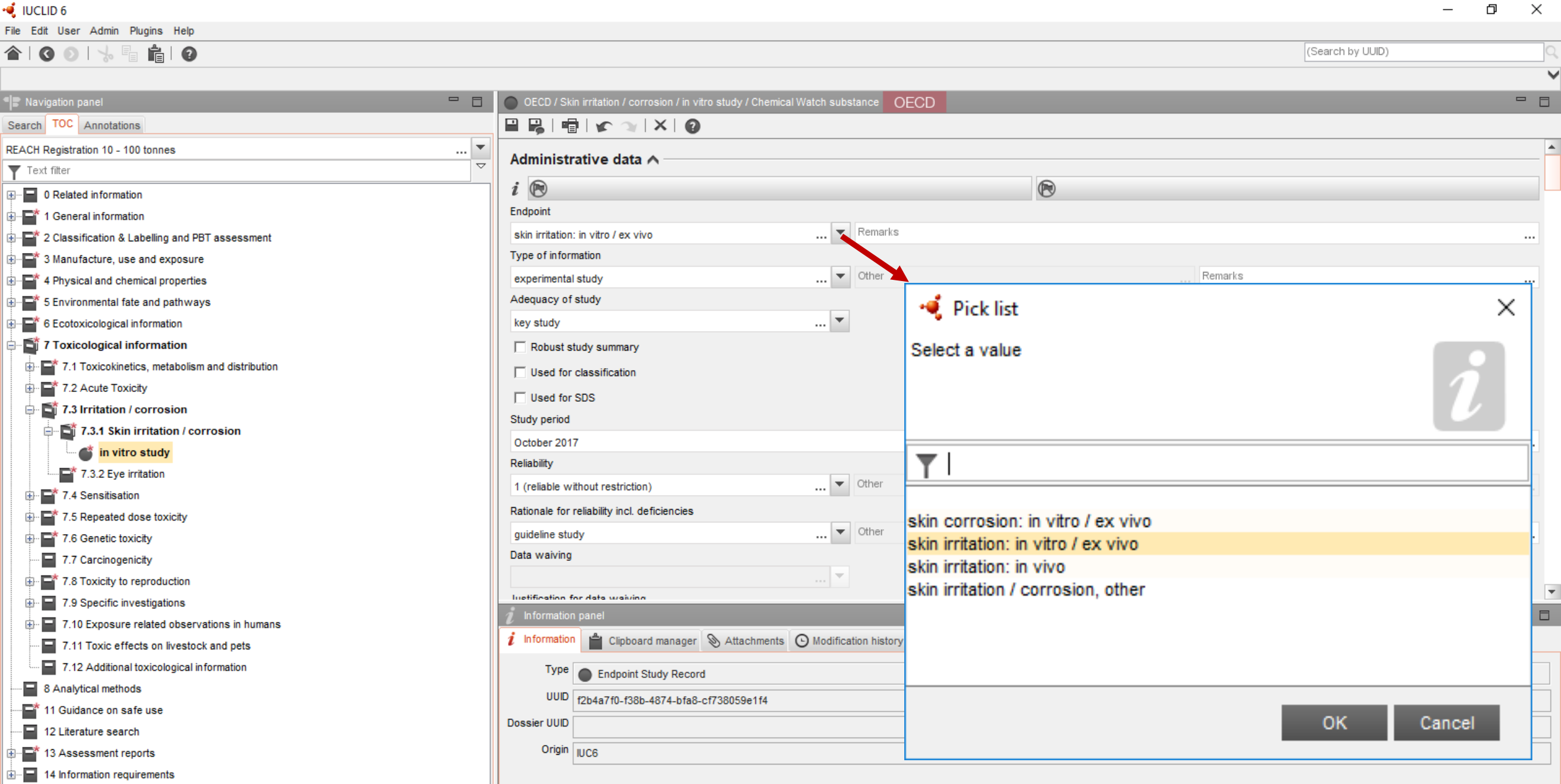
8.1.1. Skin corrosion, *in vitro*	
8.1.2. Skin irritation, *in vitro*	

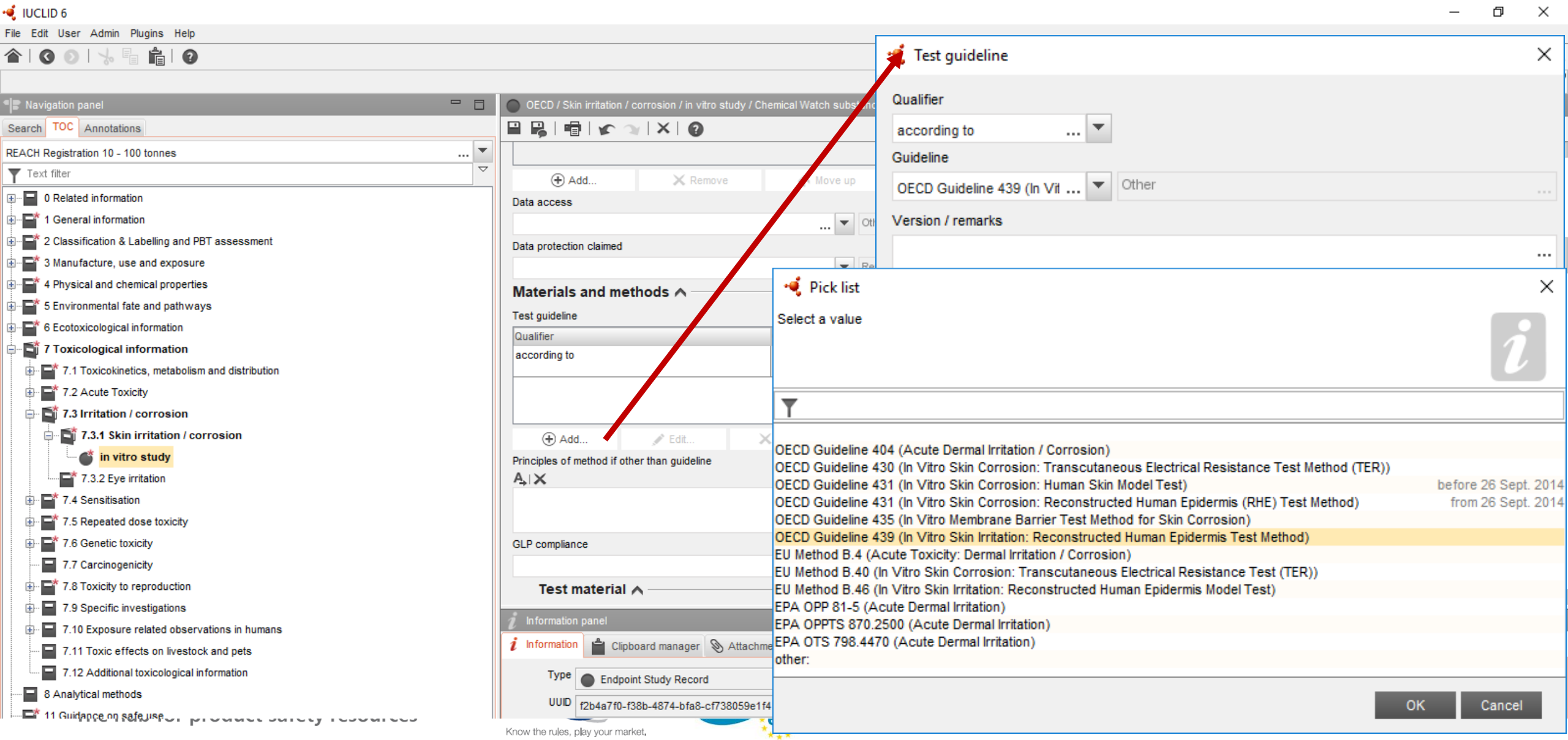
Annex VIII (< 10 t/y)

An *in vivo* study for skin corrosion/irritation shall be considered only if the *in vitro* studies under points 8.1.1 and 8.1.2 in Annex VII are not applicable, or the results of these studies are not adequate for classification and risk assessment.

Consistent IUCLID 6 Revision – *in vitro* Skin irritation / corrosion

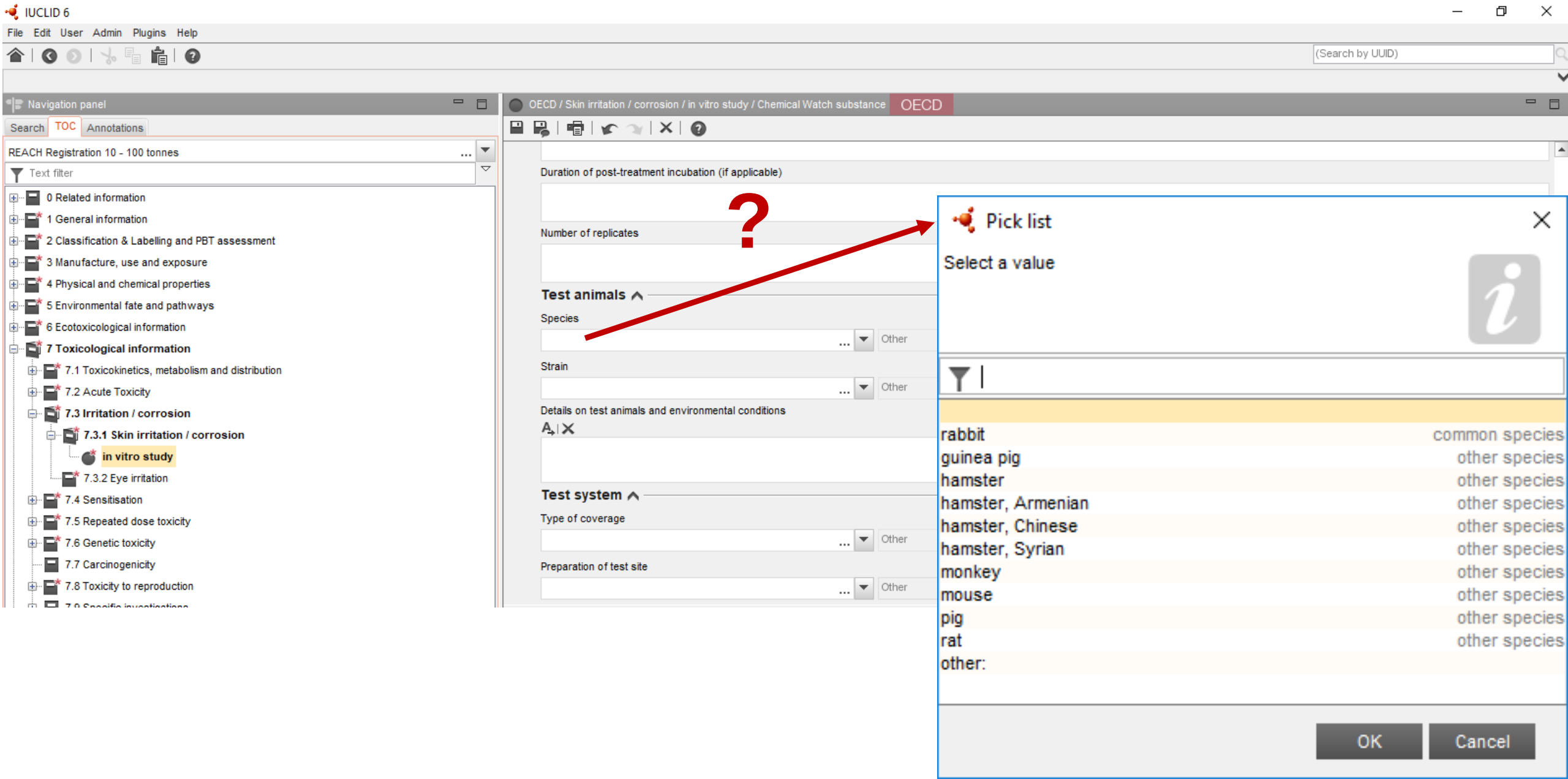


Consistent IUCLID 6 Revision – *in vitro* Skin irritation / corrosion

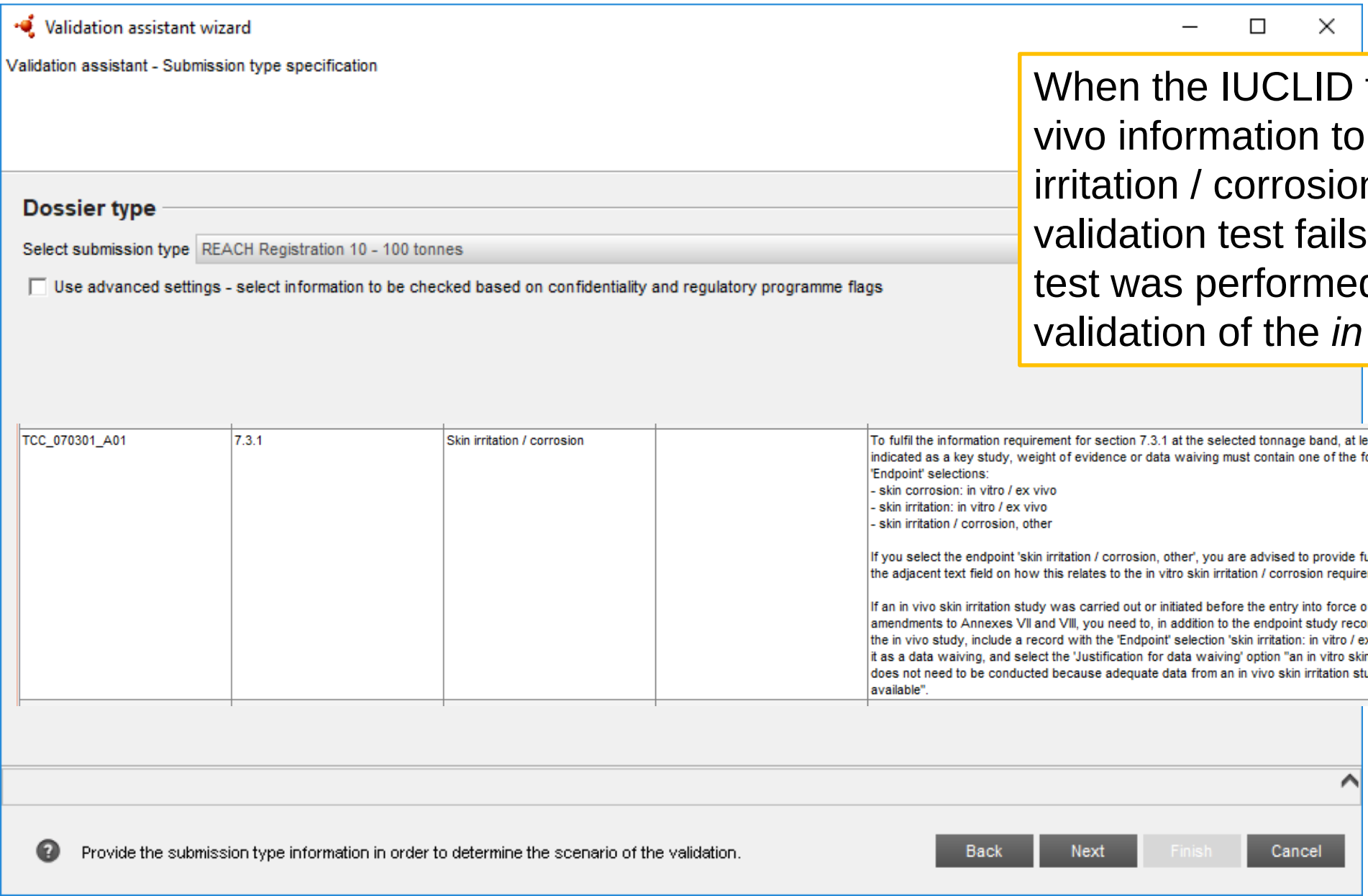


Know the rules, play your market.

Consistent IUCLID 6 Revision – *in vitro* Skin irritation / corrosion

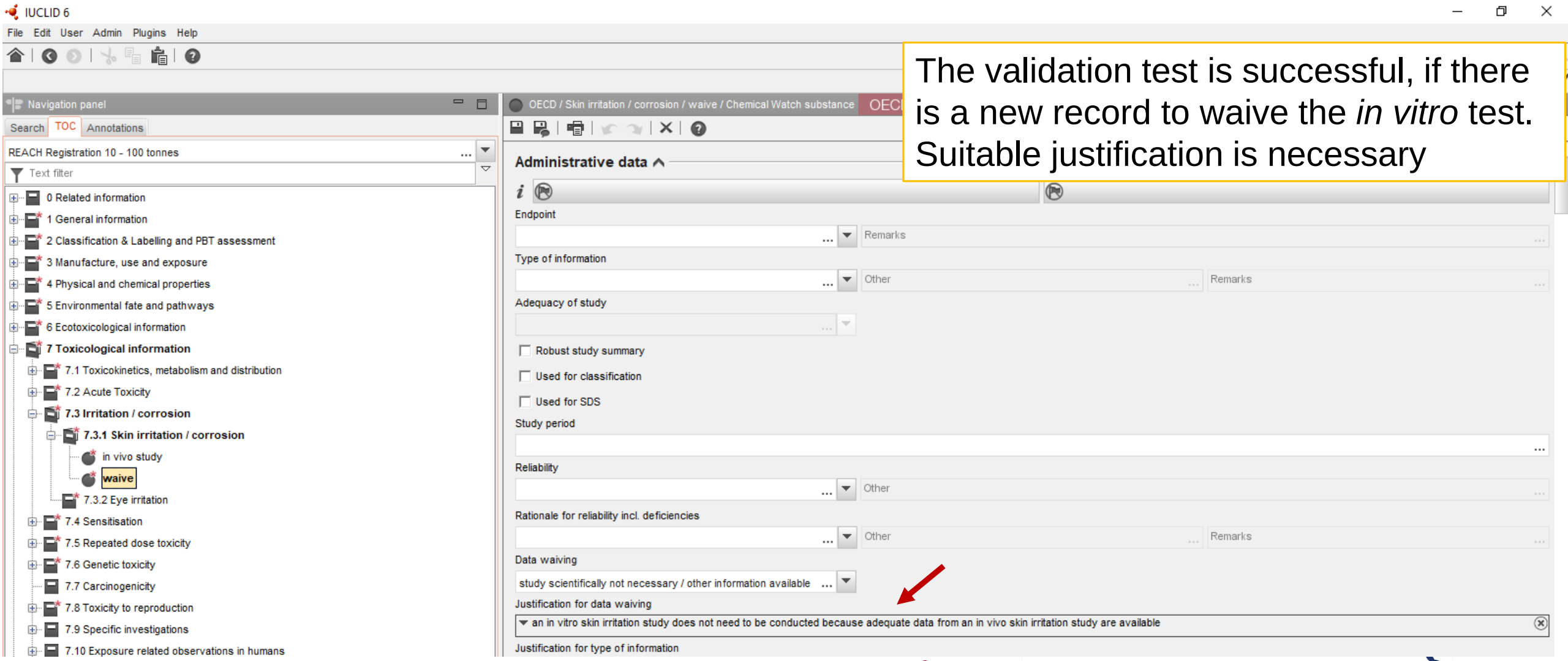


Consistent IUCLID 6 Revision – *in vitro* Skin irritation / corrosion



When the IUCLID file contains only in vivo information to cover the skin irritation / corrosion endpoint, the validation test fails, even if the submitted test was performed much before the validation of the *in vitro* tests

Consistent IUCLID 6 Revision – Waiving *in vitro* when *in vivo* exists



The validation test is successful, if there is a new record to waive the *in vitro* test. Suitable justification is necessary

What do we need?

Skin irritation	
Not irritant	
Category 2 – H315 Causes skin irritation	
Category 2 – H315 Causes skin irritation	
Category 1 – H314 Causes severe skin burns and eye damage	

What do we need?

Skin irritation	Eye irritation
Not irritant	Not irritant
Category 2 – H315 Causes skin irritation	Category 2 - H319 Causes serious eye irritation
Category 2 – H315 Causes skin irritation	Category 1- H318 Causes serious eye damage
Category 1 – H314 Causes severe skin burns and eye damage	

CLASS 8 CORROSIVE SUBSTANCES
Category 1A / 1B / 1C (Packing group I, II, III)
https://adrbook.com/en/2017/ADR/2.2.8

What do we need?

Skin irritation	Eye irritation
Not irritant	Not irritant
Category 2 – H315 Causes skin irritation	Category 2 - H319 Causes serious eye irritation
Category 2 – H315 Causes skin irritation	Category 1- H318 Causes serious eye damage
Category 1 – H314 Causes severe skin burns and eye damage	

CLASS 8 CORROSIVE SUBSTANCES
Category 1A / 1B / 1C (Packing group I, II, III)
https://adrbook.com/en/2017/ADR/2.2.8

Classified substances > 10 t/y
Hazard Assessment Conclusion for the Exposure Scenarios
Low / Medium / High
(no threshold derived)

ECHA
EUROPEAN CHEMICALS AGENCY

How to undertake a qualitative human health assessment and document it in a chemical safety report
Practical Guide 15

What's next?

We have a set of in vitro tests for the definition of the skin irritation potential

We need to fulfil some defined requirements

Section R.7.2
Skin corrosion/irritation, serious eye damage irritation and respiratory tract corrosion/irritation

Figure R.7.2–2

https://echa.europa.eu/documents/10162/13632/information_requirements_r7a_en.pdf

Testing and assessment strategy

physico-chemical properties:

1c. Is the pH of the substance ≤ 2.0 ≥ 11.5?[a]

1d. Are there other physical or chemical properties that indicate that the substance is corrosive/irritant?

Measure of many physical and chemical properties is mandatory for any dossier: do it immediately to “get to know” your substance

Testing and assessment strategy

Existing human data:

2. Are there adequate existing human data which provide evidence that the substance is a corrosive or irritant?

- Occupational exposure
  - (consumer exposure)

Testing and assessment strategy

Existing animal data:

3. Are there data from existing studies *on corrosion and irritation* in laboratory animals, which provide sound conclusive evidence that the substance is a corrosive, irritant or non-irritant?

Analysis of existing in vivo studies for one substance (From the ECHA public database)

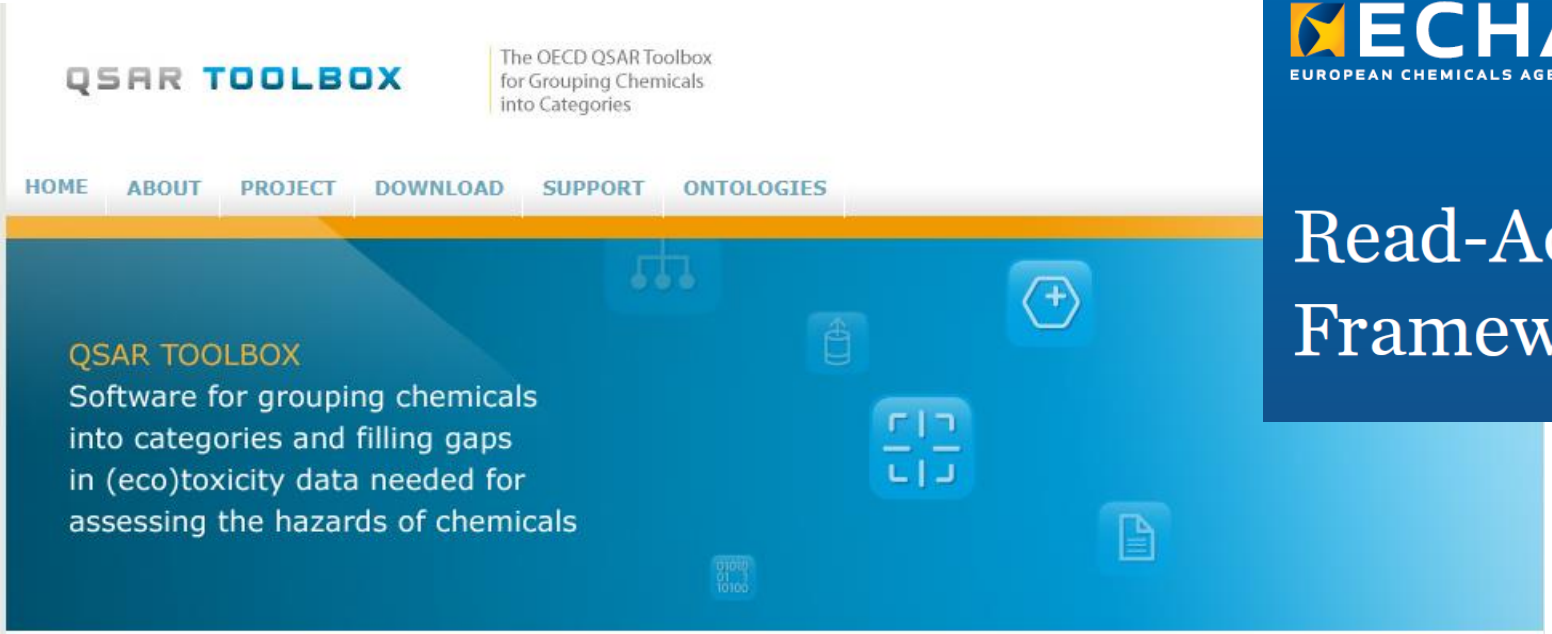
Year	Method	Species	Klimisch score	Results
1988	OECD 404	Rabbit	1	Slightly irritant
1977	24h exposure	Rabbit	3	Corrosive
1962	review article	Guinea Pig	4	Irritant
1981	24h exposure	Rabbit	3	Corrosive
1972	24h exposure	Rabbit	4	Irritant
1977	corrosivity screen	Rabbit	4	Not corrosive

ALTEX. 2016;33(2):123-34.
doi: 10.14573/altex.

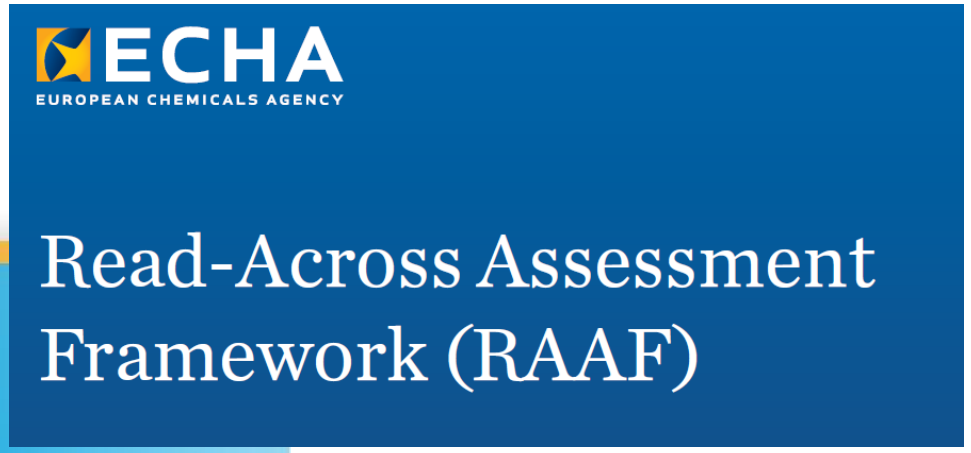
Testing and assessment strategy

Existing/new (Q)SAR data and read-across:

5a/b. Are there structurally related substances (suitable “read-across” or grouping), (…), or do suitable (Q)SAR methods indicate corrosion /irritant /not irritant potential of the substance?



www.qsartoolbox.org



https://echa.europa.eu/documents/10162/13628/raaf_en.pdf

Testing and assessment strategy

Existing in vitro data:

…

6c: Are there data from (a) non-validated suitable in vitro test(s), which provide sound conclusive evidence that the substance is corrosive/ irritant?

www.estiv2018.org

Chapter R.7a: Endpoint specific guidance

Version 5.0 – December 2016

R.7.2 Skin corrosion/irritation, serious eye damage/eye irritation …

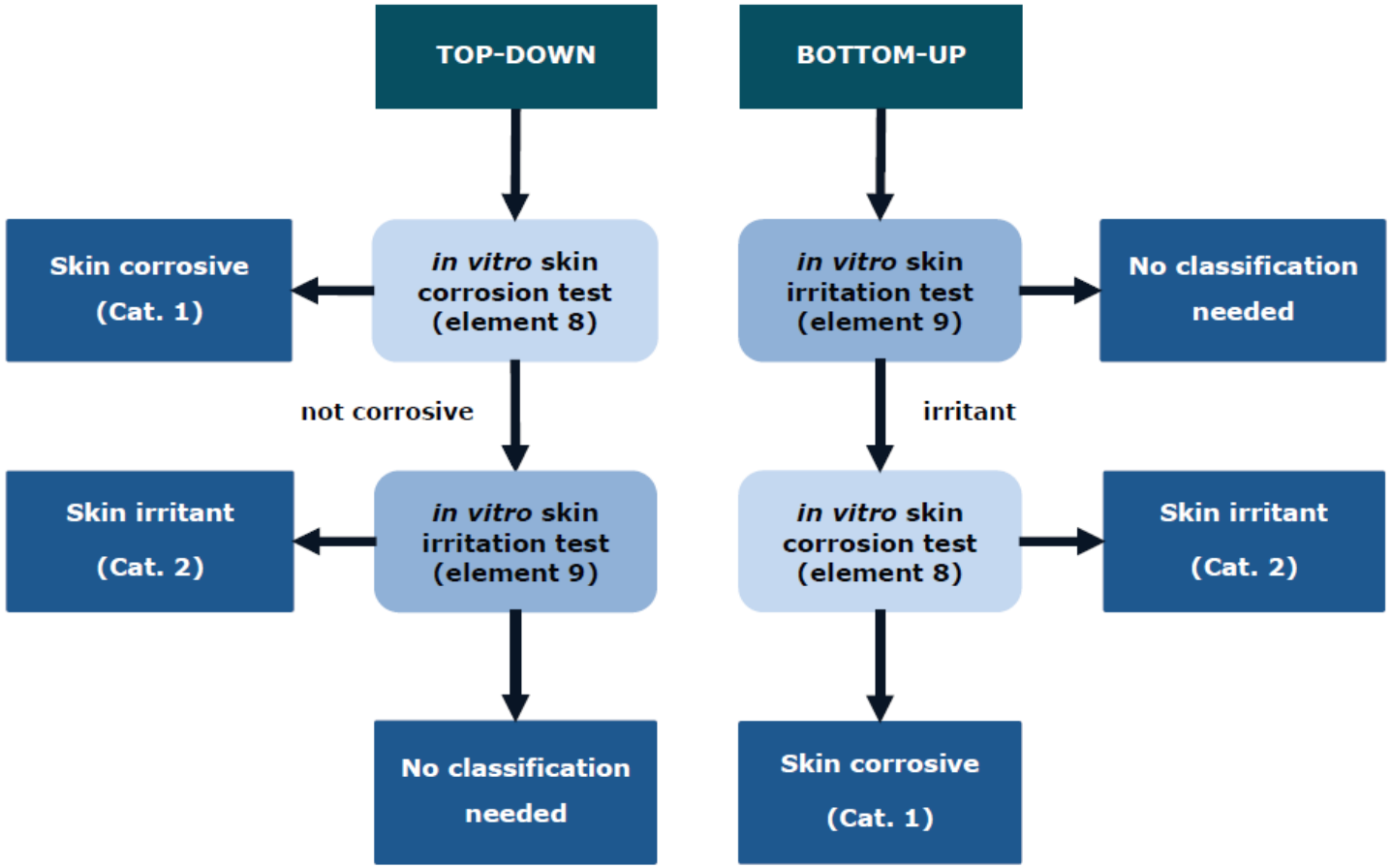


Figure R.7.2–3
Schematic presentation of Top-down and Bottom-up approaches for Skin Corrosion/irritation

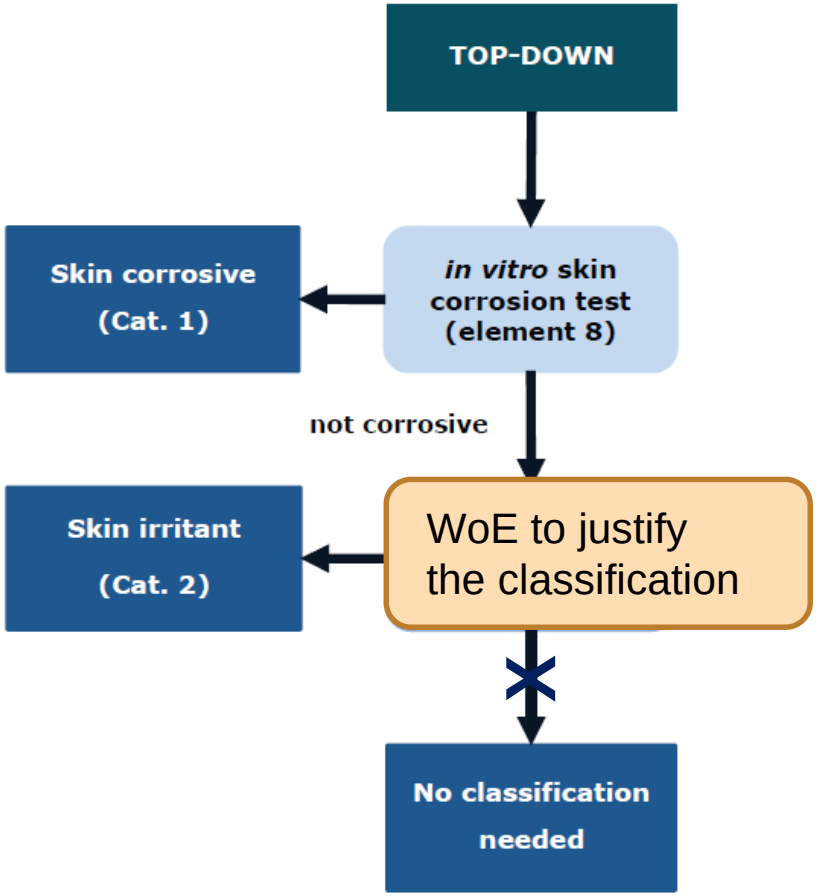
https://echa.europa.eu/documents/10162/13632/information_requirements_r7a_en.pdf

IATA for skin irritation in practice

The preliminary assessment of the substance can foresee the possible hazard very often. This defines whether to apply the Bottom-up or Top-down approach and usually one experiment is enough to get to a conclusion.
Main parameters to select the starting test are:

- pH – how much the substance is far from skin natural pH (about 5)
- MW and Kow – low interaction at high values
- Stability
- Other chemical properties (surfactant, chelating agent, and so on)
- Existing test on eye irritation
- Other existing toxicity tests
- QSAR modelling
- Anything else that may contribute to the general knowledge of the substance properties

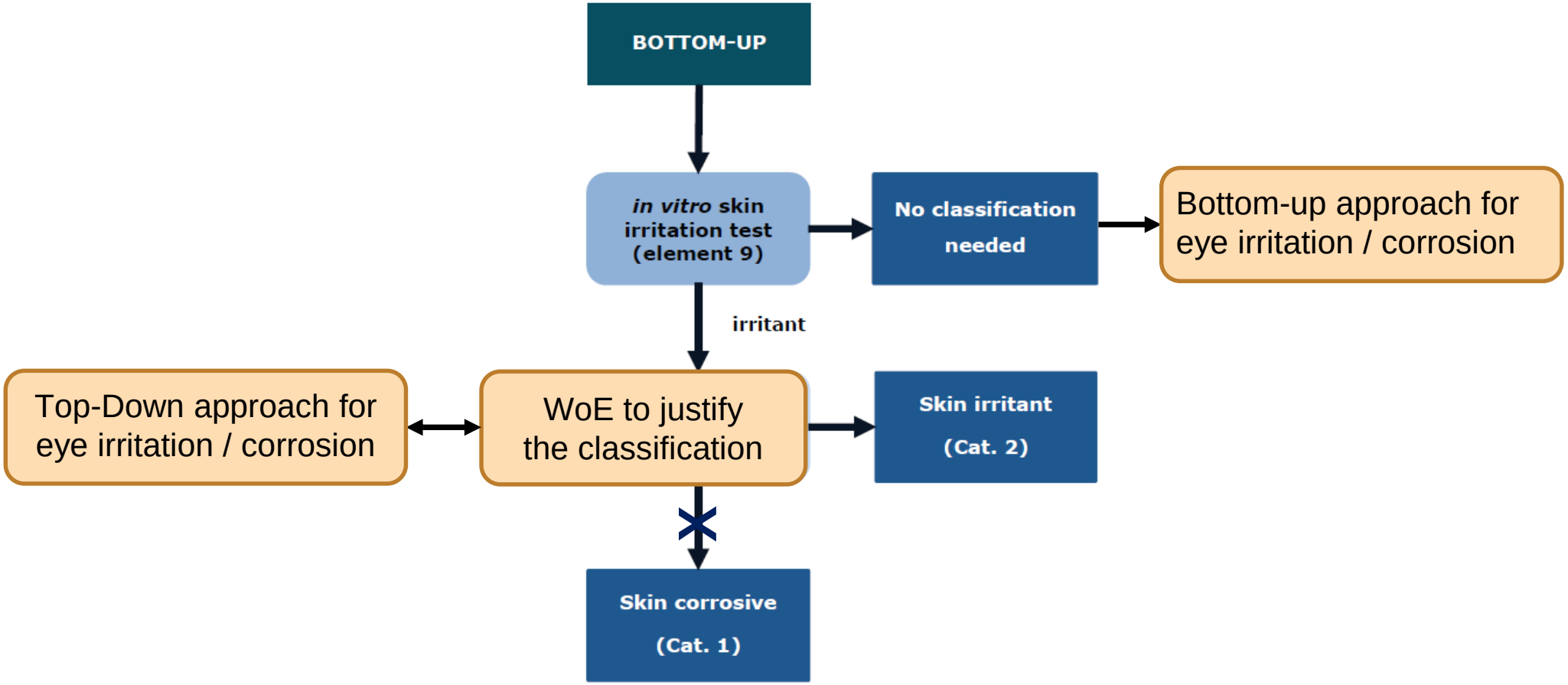
Typical top-down approach



Very often, the corrosivity hazard is well known and the test is performed to assigned the correct packing group for transport (ADR), according to OECD Guidance document 203 on IATA.

The preliminary general assessment may also generate an alert. In this case the test is performed to discriminate between Cat. 1 and 2. Confirmation of Category 2 derives from other source of information

Typical Bottom-up approach



Conclusion

- Start from the regulatory framework
- There is no unique instructions and even simple tiered strategy should be tailored to the specific substance and the specific use
- Assessment of the substance should be performed globally, not endpoint by endpoint
- Interpretation of all available results, avoid just adding the conclusions from each single study report
- Look at cost and simplicity

Q&A Session

- **Please submit questions during the webinar using your chat box.**

- Any unanswered questions can be raised in the **Chemical Risk Manager** LinkedIn group following the webinar:

www.chemicalwatch.com/CRM-LI

Speakers

- **Dr Gertrude-Emilia Costin -** Gertrude-Emilia is the manager of scientific services at the Institute for In Vitro Sciences, Inc (IIVS). In her role as study director, she is in charge of a wide variety of safety and efficacy commercial studies based on in vitro test systems. Her main area of expertise is the use of in vitro test methods for the dermal safety assessment of ingredients and final formulations manufactured by the personal-care or pharmaceutical industries as well as that of products to be registered under the US Federal Insecticide, Fungicide, and Rodenticide Act, which is regulated by the US Environmental Protection Agency.

- **Dr Costanza Rovida –** Costanza is a chemist working as a scientific officer at the Center for Alternatives to Animal Testing–Europe (CAAT-Europe) and a project manager for TEAM Mastery. Convinced that in vivo methods are not the right scientific answer to our toxicological questions, from 2005 to 2008, she worked at the European Centre for the Validation of Alternative Methods, where she gained experience in the application of alternative methods in the area of skin and respiratory sensitisation.

Chair – Name – Job Title - Organisation

Thank you for attending

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ChemicalRiskManager

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A downloadable recording of this presentation (with slides) will be available shortly.

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NEXT

- **Skin Sensitisation – Free Webinar – 1 February – 4:00pm-5:30pm GMT**
- **Chemistry for the Non-Chemist – Training Webinar – 6 February – 2:00pm-4:00pm GMT**

Please note, a recording and a copy of the presentation slides will be made available to those registered in the next few days!