The Final EUTOXRISK Symposium 1

EU-ToxRisk technologies and sustainable datasets

The EU-ToxRisk Consortium

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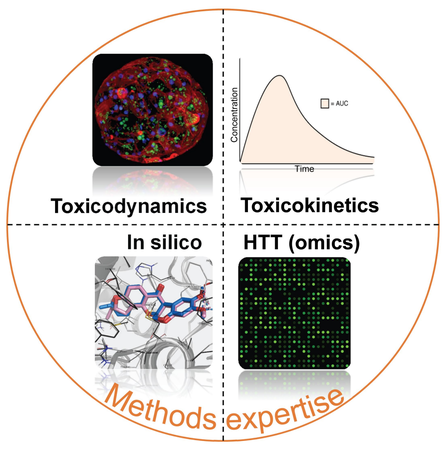
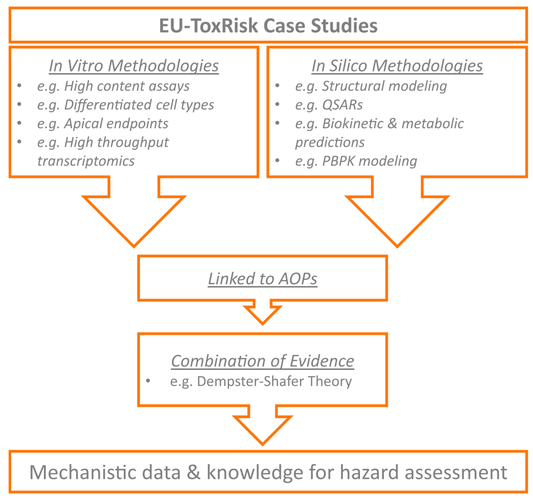


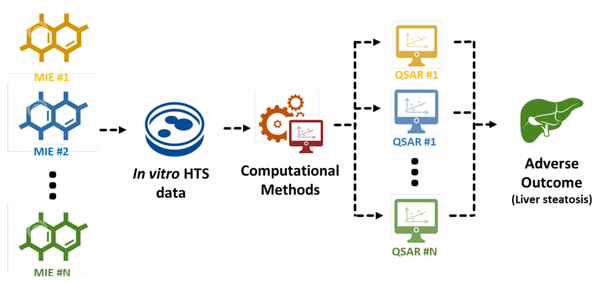
Background

EU-ToxRisk unites in silico and in vitro expertise to yield mechanistic hazard assessment by combining complementary new approach methods (NAMs) in the testing strategies. NAMs were combined and tested in multiple case studies. Linking NAM data to knowledge of adverse outcome pathways (AOPs) integrates the information for chemical safety assessment.

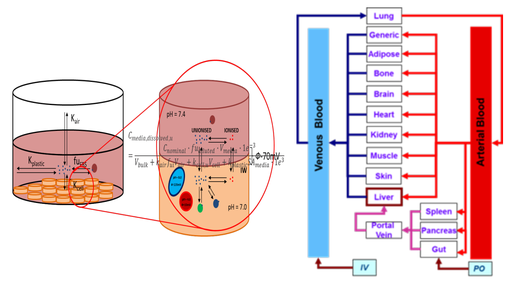


In silico

In silico methodologies, e.g.:
- **Structural modelling** – structural similarity metrics based on well-known structural fingerprints (e.g., Tanimoto index)
- **QSARs** – structural and physicochemical features associated with biological properties; EU-ToxRisk developed dozens of models covering many endpoints and biological properties
- **Biokinetic & metabolic** predictions – biokinetic models using physicochemical and/or in vitro measured data and simulating intracellular concentrations in vitro
- **PBPK modelling** – compound concentrations in target organs in vivo compared with intracellular concentrations causing toxicity in vitro -> human-equivalent in vivo doses.



EU-ToxRisk computational prediction tools (WP3) → **poster #2**



EU-ToxRisk PBPK methods (WP4) → **poster #3**

In vitro

In vitro test systems cover key **RDT endpoints** (liver, kidneys, neuronal system, lung toxicity) as well as **DART**.

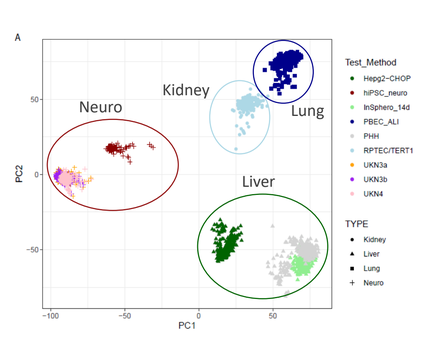
Test systems cover three levels of complexity: 1) **high-throughput** test systems; 2) **organ-specific** models; 3) **complex or disease-specific** models.; providing direct mechanistic data for the toxicological effects or endpoints of interest:
- High-throughput tests (e.g., CALUX assay and HepG2-BAC-GFP reporter assays): information on molecular signalling events and activation of cellular stress response pathways and cytotoxicity.
- Organ-specific models: information on specific endpoints such as neurite outgrowth as well as cell viability in human-relevant target organ specific test systems.
- Complex and disease-specific models: effects of chemicals in, e.g., diseased 3D liver spheroids, or on developmental processes , e.g. differentiation of mouse embryonic stem cells or the zebrafish embryo test.

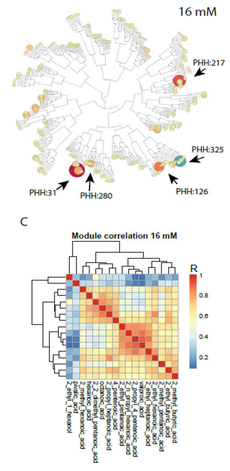
The project tested the applicability and sensitivity of the tests by **testing 19 well-described toxicants in all EU-ToxRisk assays**.

Tests address **KEs of AOPs**, including human **iPSC-derived organ-specific reporter cell lines**. In addition, **transcriptomics and biokinetic data from all test systems exposed** to the 19 toxicants were generated. Broad concentration range data allowed **PoD identification** for various measurements.

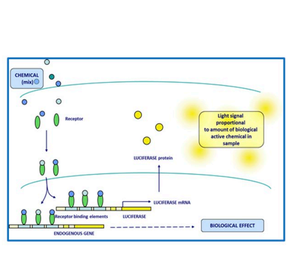
Transcriptomics of all test systems without any chemical stressors yielded insight into the make-up of the test systems.

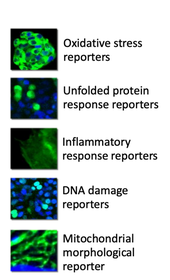
Development of innovative EU-ToxRisk in vitro test systems:
- CRISPR/Cas9 generated fluorescent protein **reporter cell lines in human iPSC** → genetically stable cell lines that can proliferate indefinitely and be differentiated into numerous target organ-specific cell lineages.
- **Dual reporter cell lines** in HepG2-BAC reporter technology
- Application of **micro-physiological systems** (MPS), including 3D organoids and multiorgan-chip technology.

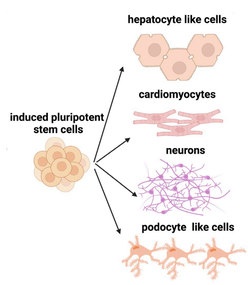




High throughput transcriptomics for hazard characterization (WP5) → **poster #4**



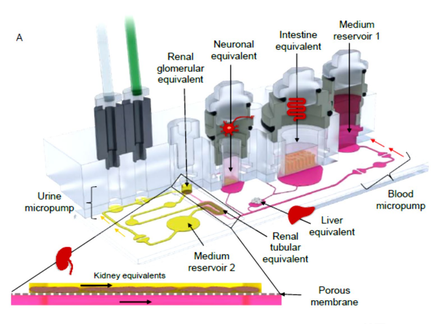




Reporter systems for MIE and KE hazard characterization (WP6) → **poster #5**

Compound	cMINC	UKN4 LUHMES	UKN3a LUHMES	UKN3b LUHMES	UKN5 PeriTox	PBEC		RPTEC/ TERT1	SH-SY5Y	
	Ratio (viability/ migration)	Ratio (viability/ neurite area)	Ratio (viability/ neurite area)	Ratio (viability/ neurite area)	Ratio (viability/ neurite area)	Ratio (viability/ proliferation)	Ratio (viability/ mitochondria)	Ratio (viability/ lactate)	Ratio (viability/ Ca2+ VDCC)	Ratio (viability/ Ca2+ basal)
Acrylamide	6.0	1.5	1.6	1.3	8.2	18.5	16.2	5.9	1.5	1.1
Carbaryl	1.0	5.5	1.5	3.4	2.4	27.0	14.3	1.0	1.5	1.2
Clofibrate	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0
Colchicine	1.1	7082.2	1.1	3.7	31.6	0.6	0.9	1.0	0.3	0.3
Hexachlorophene	1.2	0.9	1.5	2.3	1.5	NA	NA	0.4	0.7	0.0
HgCl2	2.2	1.0	1.1	0.6	1.2	1.0	0.8	NA	1.1	1.5
Ibuprofen	1.0	1.0	1.0	1.0	1.0	16.4	5.6	1.0	2.2	1.2
MPP+	1.0	52.9	1.8	1.0	1.0	61.8	87.5	1.5	1.6	0.5
Paracetamol	1.0	1.0	1.0	1.0	1.0	NA	NA	1.9	28.1	5.4
Paraquat	5.5	1.0	1.3	1.5	9.1	8.4	11.5	0.5	1.6	1.5
PCB180	5.0	0.5	0.8	0.8	1.0	1.5	1.5	NA	1.0	1.0
Rifampicin	1.0	5.5	1.2	1.5	1.0	5.0	2.4	4.3	6.8	1.8
Rotenone	4.4	153.4	11.8	9.5	79.4	37.5	600.0	5.5	2.9	2.0
Sulfisoxazole	1.0	1.0	1.0	1.0	1.0	1.2	1.1	1.0	1.0	1.0
Taxol	31.6	11.7	4.8	367.6	79.8	NA	NA	1.0	1.6	3.6
Tebuconazole	1.0	7.1	8.7	6.0	1.9	1.0	0.5	1.9	0.9	0.7
Tolbutamide	1.0	1.0	1.0	1.0	1.0	7.7	4.2	1.0	1.0	1.0
Triphenylphosphate	1.0	1.0	11.3	1.2	1.0	NA	NA	1.0	1.0	1.0
VPA	1.0	1.0	1.5	1.0	0.9	12.2	8.9	1.0	0.5	1.2

Cross-systems cell-based viability tests → **poster #6**

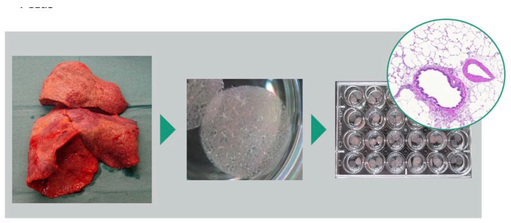


Advanced test methods → **poster #7**

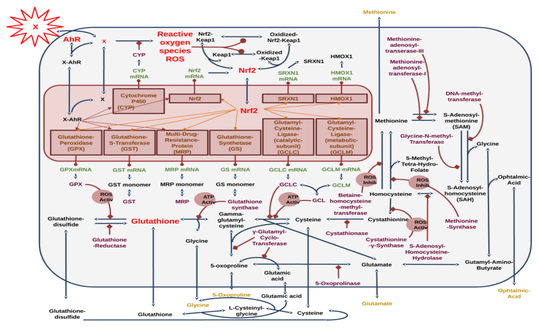
Human anchoring & qAOP

Human anchoring of NAM data:
- **anchoring KEs in AOPs** using a combination of data derived from complex human tissues ex vivo, data from human clinical exposures and from archived data from animal exposures.
- Highlighting the significance of **genetic variability and human disease** for adverse outcomes.

Development of qAOPs:
- Driving knowledge management and **data integration** for safety assessment.
- **Forecast chemical effects in humans** via in vitro and computational chemistry methods.



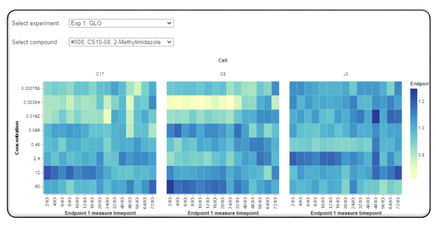
From in vitro to human translational strategies → **poster #8**



Computational methods based on qAOP → **poster #9**

Reusable data

- **BioStudies** data repository hosts **all EU-ToxRisk experimental data** and access.
- More complex data access and analysis is supported by the extended **EU-ToxRisk Knowledge Infrastructure** developed at Edelweiss Connect.
- EU-ToxRisk project data will be **FAIR and** made **public** as much as possible by April 2022.



Databasing and data sustainability for data reuse → **poster #10**

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www.eu-toxrisk.eu/