Classification of chemicals using sensitivity ratios

Stefan Scholz, Nils Klüver, Jo Nyffeler
stefan.scholz@ufz.de, Department of Ecotoxicology, Helmholtz Centre for Environmental Research – UFZ

Background

Our approach is based on the classification of compounds via their toxic ratios or sensitivity ratios. These ratios are obtained by comparing the observed toxicity in alternative test systems with the so-called predicted baseline toxicity, i.e. the intrinsic minimal toxicity (mortality, cytotoxicity) of a compound driven by its hydrophobicity. This concept was originally developed in ecotoxicology (Verhaar, 1992). However, it is applicable to any water-borne exposure system, given that the minimal or baseline toxicity is driven by the partition between water and cellular membranes. Therefore, the baseline toxicity concept is also applicable to the field of human toxicology when alternative test systems are used that are based on a water born exposure (which is the case for in vitro cellular or fish embryo test systems).
We combined data from human cellular in vitro tests and fish embryos, given that the latter is able to capture complex interactions difficult to obtain by cellular systems.

It can be used to identify compounds with a specific, reactive or uncoupling mode of action. Compounds with an observed effect well below the predicted baseline toxicity can be considered as of particular high risk for human toxicity, as it is likely that they exhibit a specific mode of action and target a pathway that is of high relevance for human health.
Note that this approach is only valid for organic chemicals and not for metalorganic chemicals, metals, polymers and mixtures. Furthermore, the baseline toxicity approach is not well described for very hydrophilic compounds. Therefore we used a cut-off of "0" for the DlipW of hydrophilic compounds. I.e. for all compounds with a DlipW below "0" the baseline toxicity was calculated using a value of "0". This was done to avoid artificially high toxic or sensitivity ratios.

Approach

Physicochemical data retrieval

COMPTOX dashboard) (https://comptox.epa.gov/dashboard/)
LSER database (https://www.ufz.de/index.php?en=31698).
ACD percepta (https://www.acdlabs.com). pka values were obtained from the commercial software

Parameter	Source	Aim
Log Kow	https://comptox.epa.gov/dashboard/)	KlipW QSAR (if no klipW available
KlipW	https://www.ufz.de/index.php?en=31698	Calculate DlipW
pkA	https://www.acdlabs.com	Calculate DlipW

Estimate baseline toxicity for neutral and disocciating compounds

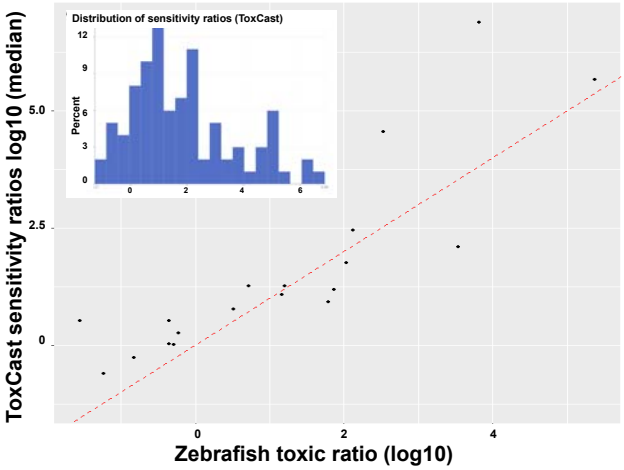
Effect data retrieval

Fish embryo toxicity database (Sobanska et al. 2018)

In vitro cellular effect data (Comptox), median values (Nyffeler 2023)

Experimental values could be obtained for 96 compounds from ToxCasts and (overlapping) 21 compounds in fish embryos

Sensitivity ratios (SR) and Toxic ratios (TR)



Relationship of ToxCast Sensitivity and zebrafish toxicity ratios (Insert: distribution of ratios for ToxCast data)

Classification scheme

- Chemicals with an SR or TR below 10 were considered as of **low risk** if their DlipW was below 4.
- Compounds with an SR or TR between 10 and 100 were considered as **medium risk** compounds.
- All with SR or TR > 100 and/or DlipW > 5 were considered as **high risk** compound.
- The classification was based on the highest **SR or TR found in any of the considered test system**.
- Inorganic and metalorganic compound could not be classified by this approach.

96 of the 150 Designathon compounds could be classified

Class	Number of compounds
High	52
Low	17
Medium	27
Medium or High	4
Not classified	52

Top10 "high risk" compounds

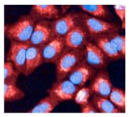
CAS no provided by EPAA	CAS No of neutral form	Compound name	Mode of action/use group	Maximum sensitivity or toxic ratio obtained
630-60-4	630-60-4	ouabain	Cardio Na+/K+ ATPase inhibitor	7636358
12427-38-2	12427-38-2	maneb (ISO); manganese ethylenebis(dithiocarbamate) (polymeric)	Extracellular matrix crosslinking inhibitor	452779
22839-47-0	22839-47-0	Aspartame	Artificial sweetener	94817
33996-33-7	33996-33-7	1-acetyl-4-hydroxy-L-proline	Anti-inflammatory drug	88896
569-64-2	569-64-2	malachite green hydrochloride	Dye with potential carcinogenicity	79759
50-70-4	50-70-4	D-sorbitol	Sweetener	58195
2451-62-9	2451-62-9	1,3,5-tris(oxiranylmethyl)-1,3,5-triazine-2,4,6(1H,3H,5H)-trione; TGIC	Not identified	40540
64-86-8	64-86-8	colchicine	Microtubuline inhibition	34907
77182-82-2	51276-47-2	glufosinate ammonium (ISO); ammonium 2-amino-4-(hydroxymethylphosphinyl)butyrate	Herbicide	20412
135-51-3	148-75-4	Disodium 3-hydroxynaphthalene-2,7-disulphonate	Not identified	6712

Conclusions

Sensitivity ratios can be used to identify hazard or risk classes for a high number of chemicals using data from in vitro or alternative testing approaches. Among the compounds classified as "High" compounds with known mode of action and high toxicity were found, such as ouabain and colchicine. A detailed assessment is needed to understand why some other compounds considered so far as of low toxic risk were also classified as high.

Outlook

We propose that the sensitivity ratio approach can be extended to provide more sensitive and mechanistic endpoints using high content and/or high throughput approaches such as cell painting or image/video-based assessment in zebrafish embryos (Nyffeler 2023, Nöth 2023).



References

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