Identification of Reactive Compounds from Structure-Based Methods

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INTRODUCTION

- The REACH legislation has led to considerable interest in QSAR modelling for the prediction of environmental toxicity and fate.
- Toxicity datasets frequently consist of groups of compounds acting via a number of differing mechanisms of action.
- Chemicals with excessive toxicity above narcosis may covalently bind to proteins
- Identification of chemicals with excess toxicity is essential for mechanistically based modelling (QSAR or read across) of environmental effects

AIMS

- The aim of this study was to develop a SMARTS matching algorithm capable of identifying 'reactive' chemicals. This algorithm was based on the work of Aptula and Roberts.[1]
- The algorithm was assessed using a dataset of fish toxicity with previously assigned mechanisms of action.[2]

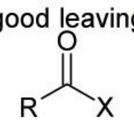
METHODS

Programming

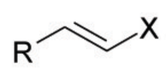
- All programming was carried out using the PERL programming language, making use of the SMARTS matching functionality of the PerlMol library.
- The ToxClassifier algorithm takes a SMILES file as input, returning several CSV files as output containing the classified chemicals
- Chemical classes were defined as SMARTS strings in an editable input file
- Five mechanisms of electrophilic action were defined as described by Aptula and Roberts in relation to skin sensitisation. These are also considered important in fish toxicity.

Mechanisms of Action Identified by the ToxClassifier Algorithm

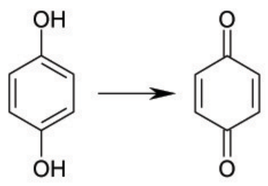
Acylation Chemicals containing a carbonyl group with a good leaving group attached. Where X = halogen or other sufficiently acidic group.



Michael Acceptors α, β-unsaturated chemicals where X = C(=O)H, C(=O)R, C(=O)OR, CN etc

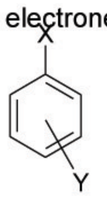


Pro-Michael Acceptors (included in the Michael class) Chemicals capable of being metabolised to Michael acceptors e.g.

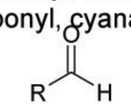


SN2 Aliphatic chemicals with a displaceable electronegative atom e.g. X = halogen.

SN2 Aromatic Activated aromatic chemicals with displaceable electronegative atoms e.g. X = halogen, Y = (two or more of) NO2, CN, C(=O)H, CF3 etc



Schiff's Base formation Chemicals with a reactive carbonyl such as aliphatic aldehydes, some ketones (not mono-ketones). C-nitroso, thio-carbonyl, cyanates act analogously



Assessment of ToxClassifier Algorithm

- The ToxClassifier algorithm was used to predict mechanisms of action for the fathead minnow database. Mechanisms had been defined for 371 chemicals previously by Russom et al[2]
- For the purposes of this study, the fathead minnow dataset was divided into narcotic (non-reactive) chemicals and those acting by electrophilic mechanisms. Other compounds acting by specific mechanisms i.e. AChE inhibition were not considered.

RESULTS

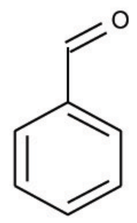
- The ToxClassifier algorithm's ability to identify electrophilic chemicals was compared to the classification presented by Russom et al[2]
- The classification by Russom et al[2] was based on empirical analysis of FATS data and investigation of excess toxicity from baseline narcosis models
- Chemicals not identified by the algorithm as being electrophiles were assumed to be narcotics
- Table 1 summarises the results from the ToxClassifier algorithm

	Electrophilic Reactive	Narcosis
Russom et al	96	275
Number **correctly** identified by ToxClassifier algorithm	50	221
Number **incorrectly** identified by ToxClassifier algorithm	46	54

Table 1: Number of correctly classified chemicals compared to Russom et al[2]

DISCUSSION

- The results summarised in Table 1 indicate the relatively poor performance of the ToxClassifier algorithm in identifying electrophiles in as compared to the classification by Russom et al[2]
- Much can be learnt regarding the prediction of reactive compounds from the assessment of the incorrect prediction. For instance, 17 chemicals identified by Russom et al[2] as reactive are aromatic aldehydes



- Aromatic aldehydes are not reactive in skin sensitisation tests. Therefore no rule exists for their identification and it is not reported by Aptula and Roberts[1]
- In addition, seven chemicals were identified as being pro-electrophilic e.g. by oxidation of a primary or secondary alcohol to an aldehyde or ketone. This is known to produces a Michael acceptor when conjugated to an alkene or alkyne
- The pro-electrophilic mechanism does exist in the skin data but is not captured in the rules developed by Aptula and Roberts[1]
- A further 20 chemicals are reported to be reactive by Russom et al[2] but were not identified by the algorithm. These chemicals were mainly un-conjugated alkene and alkyne species which are thought not to be reactive in the LLNA skin assay

CONCLUSIONS

- This study has developed an algorithm able to identify differing electrophilic mechanisms thought to be responsible for covalent based toxicity
- Rules derived from analysis of data from the skin sensitisation assay
- The study has shown that some covalent reaction mechanisms are similar for different endpoints
- However, this study has also highlighted the importance of identifying endpoint specific rules for covalently reactive chemicals

AKNOWLEDGMENT

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References

1. Aptula A.O., D.W. Roberts., *Chem. Res. Tox.*, **19**, 1097 (2006)
2. Russom, C.L., et al, *Environ. Tox. Chem.*, **16**, 948 (1997)
3. PerlMol Chemical Library, written by Ivan Tubert-Brohman, freely available from: http://www.perlmol.org/