Integrating Toxicity Testing and Chemical Analyses to Identify Toxic Chemicals in Our Environment

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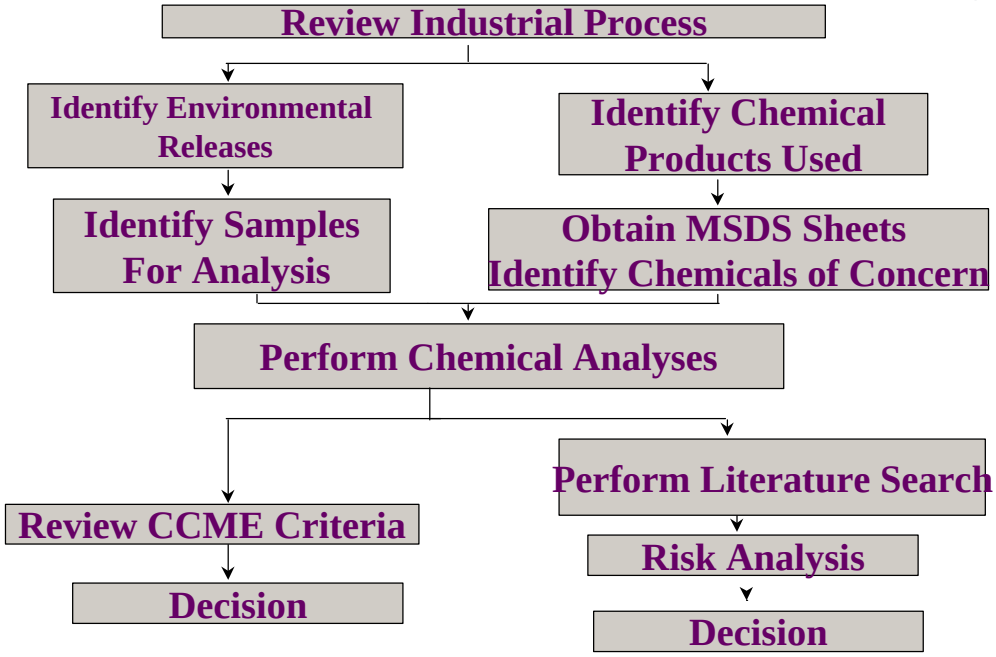
What is the Operating Logic?

- Assume that priority pollutants represent the universe of “toxic” chemicals
- Assume that GC/MS open scans will identify any other chemicals that the consultant might miss and should be concerned about
- Assume that metal scans will complete the picture, i.e. go from 13 to 25 metals or more
- Everything I need to know or be concerned about will be covered off with these tests
- This is the way everybody does it!

Conventional Risk Assessment



Reality Check

- **How many organic compounds are there?**
- **How many of them are toxic?**
- **Can these toxic organic chemicals be detected by priority pollutant and/or GC/MS open scans?**
- **Can industrial chemicals undergo chemical transformations to more toxic and mobile endproducts and will priority pollutant and/or GC/MS scans identify these?**

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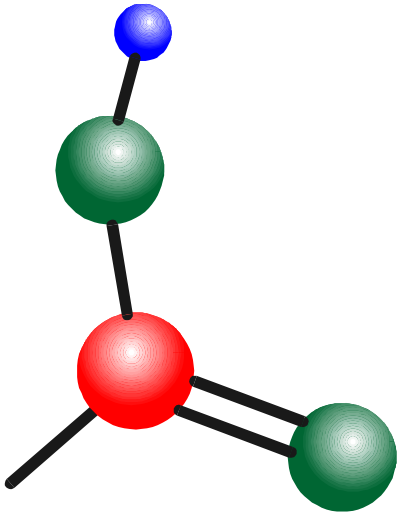
How Many Organic Chemicals Are There?

- Not sure, perhaps Billions or more
- Ask a chemist how many chemicals you can make with the periodic table and carbon?
- ACS lists over 15 million
- Approximately 500,000 added annually to ACS list

How Many Chemicals Are Toxic?

- RTECs lists about 150,000. Not free
- Chemicals added as more data is gathered
- Aquatic and mammalian databases now available thru internet (Ecotox, Toxnet, Aquatox, Terratox, Phytotox). Some fees apply
- Private data bases available but expensive
- May be difficult to find information needed and search may be expensive and not fruitful
- Toxicity values for same species may span two order of magnitude

What Is a QSAR?



- Quantitative Structure Activity Relationship (QSAR)
- Used when no toxicological data on a specific chemical can be found
- Assumes that toxicity is due to the presence of certain chemical functional groups
- These functional groups are identified within an observed chemical (GC/MS) and their contributing toxicities added to give a predicted value
- Makes certain mechanistic assumptions
- Software is available from various sources, e.g. Rutgers Medical School

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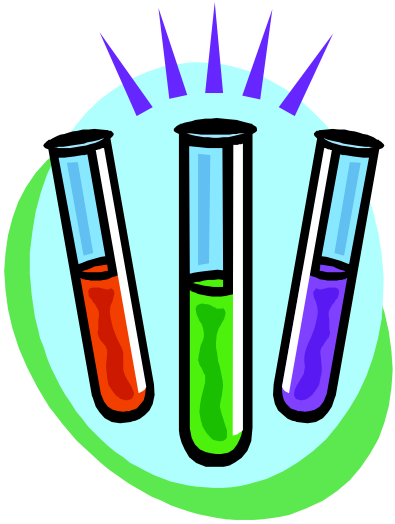
How Reliable are QSARs?



- Can be reliable if mechanism is correct and thoroughly understood
- Can be out to lunch
- Biggest problem is chemical isomerization
- Chemical isomerization can result in toxicities differing by two or more orders of magnitude, e.g. dimethyl - quinolines
- Can severly over estimate or under estimate toxicity

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If GC/MS can't measure all organic chemicals, is there another magic box that can?

- **No!** There is no current instrument where you can place the sample in one end and the description of everything in the sample comes out the other
- Need myriad of techniques including: derivatization GC/EI-MS, GC/CI-MS, LC/MS , LC/MS/MS/MS (thermospray, electrospray, API, FAB), GC/HRMS, GC/FTIR, MALDI/TOF, etc. **All these techniques have strengths and weaknesses and can be expensive**

Let's Take Stock

- Priority pollutant scans and GC/MS scans provide limited data
- A myriad of other techniques may be required to gather all the chemical information I need to make an assessment
- Even if I do get the data I need, curent toxicological databases and QSARs may be inadequate to allow for interpretation. Furthermore, significance of a finding may not be realized
- Use of the conventional approach may lead to future liabilities for client

What Can Be Done To Improve the Situation?

- Need a different logic
- Need to incorporate toxicity tests with chemical analyses
- Need procedures that integrate physical/chemical manipulation of samples, toxicity testing and chemical analyses to isolate, identify and confirm causative agent(s) for toxicity, i.e. Toxicity Identification Evaluation [TIE]

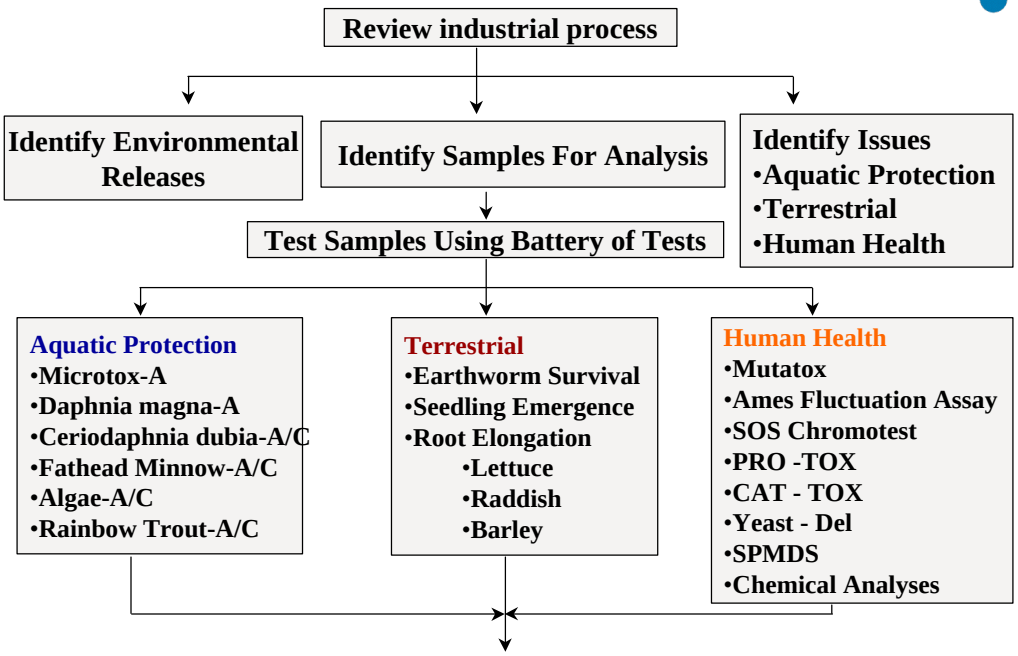
What Is This Going to Accomplish

- Determine if we have a problem
- Through the application of TIE procedures will allow us to isolate the toxicants
- Tell us what analytical procedures we need to apply in order to identify and confirm the toxicants
- Provide relevant toxicological endpoints which we can use to set criteria, e.g. NOECs, LOECs, LC_{50}'s, etc.
- Criteria include permit writing, water quality guidelines or cleanup standards

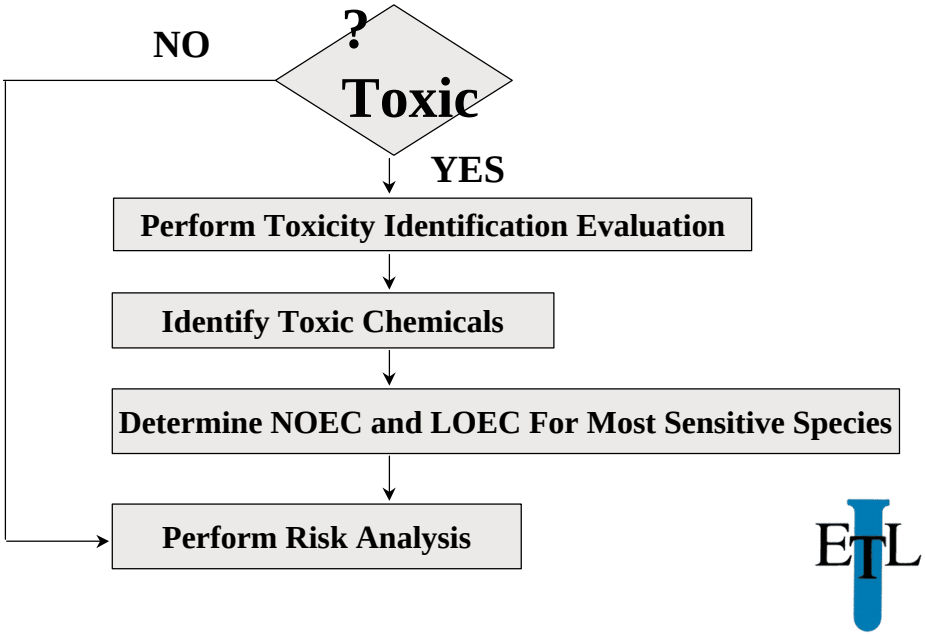
What Toxicity Tests Do I Ask For?

- What are the issues?
- Aquatic protection:
- Fish bioassay
- Invertebrates
- Bacteria
- Algae
- Terrestrial protection:
- Earthworm survival, Springtails
- Seedling emergence and root elongation
- Human health
- Genotoxic tests (Ames Test, SOS Chromotest, Mutatox, Stress Gene Assays [Pro-Tox, Cat-Tox)

Toxicity Approach For Risk Assessment



Toxicity Approach For Risk Assessment



Integrating Toxicity and Chemistry

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- **Toxicity Identification Evaluation**
- **A sample, observed to be toxic, is subjected to a logical set of physical and chemical manipulations, and the strength of toxicity before and after each treatment is used to identify toxic fractions and guide chemical analyses.**
- Identification of toxicants is efficient because chemical analyses are done only on those fractions showing toxicity, and only for the types of chemicals likely to be found
- **A site specific study conducted in a stepwise process designed to isolate, identify and confirm the causative agent(s) of toxicity [acute and/or chronic]**

Toxicity Identification Evaluation

Salient Features

- pH adjustment/aeration
- pH adjustment/filtration
- pH adjustment/solid phase extraction
- EDTA chelation
- Graduated pH test
- Oxidant reduction
- Piperonyl butoxide addition
- *Ulva Lactuca* addition
- Zeolite addition
- Cation exchange resin addition

Examples

North Saskachewan River Study

STUDY DESIGN

- **Samples of ambient water, bottom sediment and suspended sediments collected from 9-sites along 960 km reach of river**
- **Toxicity testing (*P. redivivus* and *S. typhimurium*, Ames Test) and chemical analyses for priority pollutants performed**
- **Ongley et al. (1988) Environmental Quality 17: 391 - 401**

FINDINGS

- **Toxicity associated most with suspended sediments**
- **Toxicity not correlated with priority pollutants**
- **Finding questioned water monitoring programs at the time, i.e. priority pollutant monitoring of ambient water only**

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Suncor Heavy Oil Spill

- In January, 1982 a fire and series of equipment failures resulted in the release of 50-100 tons of partially cracked bitumen to the Athabasca river in Northern Alberta
- **Preoccupation with PAHs and BTEX during investigation and evidence collection for litigation and risk assessment**
- **Toxicity approach applied to see if preoccupation was justified**
- Birkholz et al (1987). Oil in Freshwater: Chemistry, Biology, Countermeasure Technology, Eds. J.H. Vandermeulen and S.E. Hrudey, Pergamon Press, 42-57

Major Components of Toxic strong-base PANH Fraction obtained from Cracked Heavy Oil

Findings

- 2- to 5-ringed aromatic nitrogen heterocycles
- predominated by alkyl quinolines
- Plenty of mammalian toxicological information but little aquatic tox data

Conclusion

- Preoccupation with PAHs during intial investigation not justified
- Initial Water analyses after accident revealed major components were alkyl quinolines however little was done with data
- The TIE revealed that the alkyl quinolines should have been focus of risk assessment

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Canada Creosote Site

Signifigance of Approach

- Conventional analyses identified PAHs, however, toxicity revealed that something else that was toxic was present, i.e. PAHs could not account for all the toxicity
- Unconventional techniques such as derivatization GC/MS and LC/MS were required
- These analyses revealed that HPANH were major contributors
- Source of HPANH likely microbial degradation as these are not present in creosote
- HPANH added to priority list of chemicals for risk assessment

Take Home Message

- There is no universal method of chemical analysis.
- Numerous methods are required to detect everything
- Priority pollutant scans and GC/MS scans provide very limited information
- Toxicological data bases are often lacking and we may need to generate our own toxicological data
- We need a probe (e.g. toxicity test) to tell us what methods to use. Toxicity testing also tells us significance of data we have collected
- We can use our own toxicity data to set cleanup criteria (e.g. NOEC, LOEC.)
- Guideline lists of chemicals are meniscule to the known number of chemicals. Focus on guideline lists may result in misguided remediation with resultant future liabilities.

The End