Why is chemical activity successful as a metric of aquatic toxicity? A gedankenexperiment explains why.

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Introduction

- Gedankenexperiments ('thought' experiments) are widely used to predict and explain scientific phenomena when actual experiments are not feasible or practical
- The objective of this study is to show how the **chemical activity concept** can form testable hypotheses relevant to ecotoxicology
- We first hypothesize that chemical activity can be used to define acute lethal chemical activities (La_{50}s) for nonpolar and polar narcotics (i.e., baseline toxicants) and evaluate this hypothesis
- We then illustrate how chemical activity hypotheses can be used for designing aquatic toxicity tests

What is chemical activity?

- Chemical activity (*a*) relates to chemical potential, fugacity and concentration and quantifies the partitioning tendency of a chemical between phases
- In water, *a* is the concentration divided by the water solubility of the liquid-state chemical, i.e., $a_W = C_W / S_W$
- *a* in other phases can be estimated using partition coefficients, e.g., in lipids, $a_L = C_L / (K_{LW} \cdot S_W)$; K_{LW} = lipid-water partition coefficient
- At **equilibrium**, chemical **activities** in all phases are **equal**

The Gedankenexperiment

- Assumptions:
  - Fish are exposed to neutral organic chemicals where mass transport (uptake, internal distribution) is infinitely fast and equilibrium is reached instantaneously
  - The organic chemicals exert toxicity only by baseline narcosis in the target membrane phase
  - No biotransformation or growth
  - Sorption capacity of storage lipids (**SL**) equivalent to octanol, storage capacity of membrane lipids (**ML**) a % of that of octanol
- Water concentrations are varied from high to low
- Compare (test) hypotheses with measured data
- Required chemical properties:
  - Melting point (MP), water solubility (S_W), octanol-water partition coefficient (K_{OW}), molar volume (MV)

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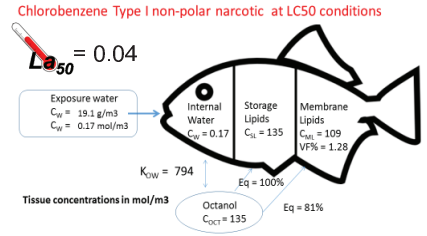
Application & Results

Table 1. Property values of nonpolar chlorobenzene (CB) and polar 2-nitrotoluene (2NT)

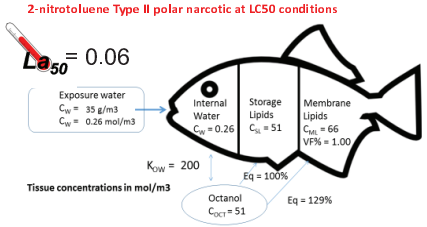
Chemical	MW g/mol	MV cm³/mol	S_W mol/m³	LC50 mol/m³	log K_{OW} log K_{SL}	log K_{ML}
CB	113	117	4.1	0.17	2.90	2.81
2NT	137	137	4.4	0.26	2.30	2.41

* Partitioning properties and LC50 based on data presented in [1] ; other property values taken from EPISUITE database. Note that both chemicals are *liquids* at ambient temperature



C_W mol/m³	a_W	C_{WW} mol/m³	C_{SL} mol/m³	C_{ML} mol/m³	Vol% Membrane	Speculative Comments
4.1	1.0	110	3290	2660	31	Saturation
1.3	0.3	35	1040	840	9.9	Immediate lethality
0.4	0.1	11	330	270	3.1	Immediate lethality
0.17	0.04	3.5	135	110	1.3	Measured LC50
0.1	0.03	1.1	104	84	1.0	Acute lethality?
0.04	0.01	0.4	33	26	0.3	Delayed lethality?
0.01	0.003	0.1	10	8.4	0.1	Chronic?
0.001	0.0003	0.03	1.0	0.8	0.01	Behavioural FX?
0.0004	0.0001	0.01	0.3	0.3	0.003	No observed FX?

Figure 1. External and internal concentrations and chemical activity at La_{50} (fish) and at various external concentrations (table). Volume fractions in membrane also shown (table)



C_W mol/m³	a_W	C_{FW} mol/m³	C_{SL} mol/m³	C_{ML} mol/m³	Vol% Membrane	Speculative Comments
4.4	1.0	38	890	1144	17	Saturation
1.4	0.3	12	280	360	5.5	Immediate lethality
0.4	0.1	4.0	90	114	1.8	Acute lethality
0.26	0.06	2.1	51	66	1.0	Measured LC50
0.1	0.03	1.2	28	36	0.6	Delayed lethality?
0.04	0.01	0.4	8.9	11	0.2	Delayed lethality?
0.01	0.003	0.1	2.8	3.6	0.06	Chronic?
0.001	0.0003	0.01	0.3	0.4	0.006	Behavioural FX?
0.0004	0.0001	0.004	0.1	0.1	0.002	No observed FX?

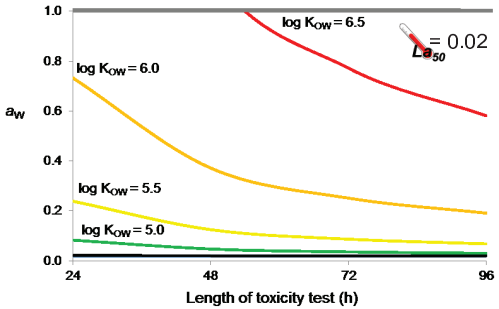
Figure 2. External and internal concentrations and chemical activity at La_{50} (fish) and at various external concentrations (table). Volume fractions in membrane also shown (table)

Discussion

- Acute lethal activities (La_{50}s) for baseline toxicants in aquatic toxicity tests tend to lie in a narrow range of 0.01 to 0.1, e.g., [2]
- Test concentrations <= 0.1 S_W are a reasonable starting point if chemicals of interest are suspected to be baseline narcotics
- Observed acute La_{50}s well below 0.01–0.1 imply a specific mode of action and/or biochemical reactivity i.e., greater potency
- More analyses are required to support the chemical activity concept

Other Considerations

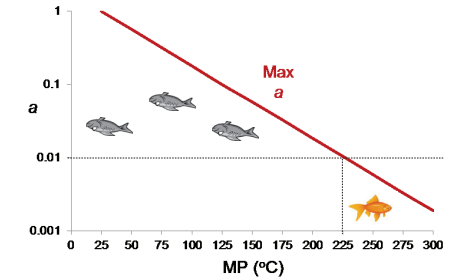
1. Are there **kinetic limitations** to achieving acute lethal effects?
   - REALITY: Equilibrium between water and fish is **not achieved** instantaneously
   - Application of a kinetic bioconcentration model allows the estimation of the external chemical activity (a_W) needed to reach the internal La_{50} in a given time



$$a_W = \frac{0.02}{1-\exp(-k_E \cdot t)}$$

KEY
As hydrophobicity ↑
a_W needs to be to exert toxicity within test period ↑
Can be assessed before starting tests

2. Can **high melting point chemicals** exert acute lethal toxicity?
   - Chemical activity of solids is calculated using *subcooled liquid* property values, which are related to solid state properties using the **Fugacity Ratio (F)**
   - The **Fugacity Ratio (F)** can be estimated as a 1st approximation from **melting point** & also equals the **maximum chemical activity** that can be exerted by the chemical



$$\log F = -0.01(MP - 25)$$

KEY
As MP ↑
F & Max *a* ↓
Not expected to exert acute toxicity
Can contribute to acute toxicity of mixture though [3]

References: [1] Vaes, W.H.J.; Ramos Urrestarazu, E.; Verhaar, H.J.M.; Hermens, J.L.M., Acute toxicity of nonpolar versus polar narcosis: Is there a difference? *Environ. Toxicol. Chem.* 1998, 17, 1380-1384. [2] Mackay, D.; Arnot, J.A.; Petkova, E.P.; Wallace, K.B.; Call, D.J.; Brooke, L.T.; Veith, G.D., The physicochemical basis of QSARs for baseline toxicity. *SAR QSAR Environ. Res.* 2009, 20, 393-414. [3] Smith, K.E.C.; Schmidt, S.N.; Dom, N.; Blust, R.; Holmstrup, M.; Mayer, P. 2013, Baseline toxic mixtures of non-toxic chemicals: "Solubility addition" increases exposure for solid hydrophobic chemicals. *Environ. Sci. Technol.* 47:2026-2033.