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Molecular Volumes and the Toxicities of Chemicals to Fish

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The biological activities of many compounds depends on their physical properties (Ferguson, 1939; Ferguson, 1951; McGowan, 1951; McGowan 1968; Ferguson and Pirie, 1948). Ferguson (1964) in a discussion on narcotics, which are examples of such compounds states "the actual process of narcosis does not involve as a first step, any chemical reaction, any breaking of covalent bonds. There are various kinds of evidence for this. Many narcotics for example, leave the body unchanged. No traces of break-down products are detected. Another piece of evidence is supplied by the very varied chemical structure of the substances which produce narcosis. Thus the same physiological reaction of all kinds of organisms is brought about by substances of such diverse chemical constitution as chloroform, ether, ethylene, cyclopropane ketones, alcohols and nitrous oxide. Even the inert gases xenon and argon have been shown to be capable of causing narcosis". In this study we show how the reported toxicities of a variety of chemicals to fish depends on the physical properties of these substances and especially their molecular volumes. Equations are given which can be used to determine which compounds in a series are physically toxic, and to predict the likely toxic concentrations of compounds.

MATERIALS AND METHODS

McGowan (1952) put forward a formula relating the concentrations of compounds, showing a given manifestation of non-selective (i.e. physical) toxicity in aqueous solutions, to the molecular volumes of the compounds measured under corresponding conditions. This treatment was based on the idea that the solution of a compound in a liquid can be considered as a two stage process. First, cavities have to be formed in the solvent to accommodate the molecules of the compound and then the molecules of the compound have to be transferred to these cavities. The work involved in this second stage does not vary greatly from solvent to solvent and is, even for a highly associated liquid like water, close to that for organic solvents. Equation (1) was suggested by these considerations.

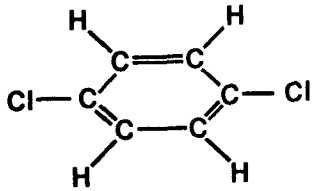
$$-\mathrm{Log}_{10}C_t + \mathrm{Log}_{10}C_B = 0.012\ V_P - E. \qquad (1)$$

where C_t are toxic concentrations in water, C_B is a constant for a given manifestation of toxicity and represents the toxic concentration in some biophase. The parachor V_P (in c.g.s units) was originally used as a measure of molecular volume but the molecular volume V_x (in S.I. units) at the absolute zero is more satisfactory and is now more often used. The term E is a correction for associated substances and compounds with a tendency to hydrogen bonding: it is zero for non-associated compounds with no tendency to take part in hydrogen bonding. Equation (1) can be rewritten with molecular volume V_x in the units $m^3\ mol^{-1}$ as shown below.

$$-\log_{10}C_t + \log_{10}C_B = 36000\ V_x - E \qquad (2)$$

Aquatic toxicologists often express concentrations as milligrams per litre but with C_t expressed in this way C_B would depend on the molecular weight of the compound and would not be a constant for a given manifestation of toxicity. The volume units used here are all cubic metres. The figures for the concentrations would be unchanged if expressed as millimols per litre but it would be unusual to express molecular volumes as litres per millimol.

Molecular volumes at the absolute zero, or characteristic volumes V_x, can be calculated by the addition of factors. Atomic values of V_x for each atom in the molecule of the compound are added together. The number of covalent bonds in the compound are counted, but the different kinds of covalent bonds are not distinguished, and this number multiplied by a factor (6.56×10^{-6} $m^3\ mol^{-1}$) is subtracted from the sum of the atomic V_x values to give the characteristic volume for the compound. A list of atomic characteristic volumes has been given (McGowan, 1978). Thevalues used below are (in $m^3\ mol^{-1}$): carbon (1.635×10^{-5}); nitrogen (1.439×10^{-5}); oxygen (1.243×10^{-5}); hydrogen (8.71×10^{-6}); chlorine (2.095×10^{-5}) and bromine (2.621×10^{-5}).



Paradichlorobenzene may be taken as an example to show how V_x values are calculated. There are six carbon atoms, ($6 \times 1.635 \times 10^{-5}$) = 9.810×10^{-5}; four hydrogen atoms, ($4 \times 8.71 \times 10^{-6}$) = 3.484×10^{-5}; two chlorine atoms, ($2 \times 2.095 \times 10^{-5}$) = 4.190×10^{-5}. The sum for these atoms is 17.484×10^{-5} and from this is subtracted that for 12 bonds, ($12 \times 6.56 \times 10^{-6}$) = 7.872×10^{-5}, to give the V_x for p-dichlorobenzene, 9.612×10^{-5} $m^3\ mol^{-1}$. The

V_x values for o- and m-dichlorobenzene would of course be the same as for p-dichlorobenzene. An equation similar to (2) has been used to estimate the solubilities of organic compounds in water and the partition coefficients of compounds between organic liquids and water (McGowan, 1954).

RESULTS AND DISCUSSION

Alexander et al. (1978) and Mayes et al. (1983) measured the median lethal dose of hydrocarbons for fathead minnows (Pimephales promelis) after 96 h at 12°C. These measured toxicities are compared in Table 1 with toxicities calculated from molecular volumes using an equation (3) based on equation (2).

$$\log_{10} C_t = -36000\ V_x + 2.1 \qquad (3)$$

In equation (3), the value + 2.1 has been used for $\log_{10} C_B$, and zero for E. This clearly is not the case, and while the agreement between the observed and calculated values in Table 1 is good in most cases, some compounds which contain polar groups show poor agreement. Thus the observed toxic concentrations for ethanediol and 2,2'-dihydroxydiethylamine (diethanolamine) are much higher than those calculated from equation (3) because the interaction of these two compounds with water have been neglected. The figures in brackets on the table allow for two interacting groups which interact with water, and are in reasonable agreement with the observed values. The interaction for each alcohol, ester and carbonyl group requires E = 1.2 in equation (2) (McGowan, 1968; McGowan, 1954). 1-Chloro-2,3-epoxy-propane is more toxic than would be expected from equation (3). The toxicity probably arises from this being a chemically reactive compound. Phenols and phenolic ethers give lower interactions than the groups mentioned above and a value for E equal to 0.6 has been suggested by McGowan (1968, 1954) The interactions may be weak and, in Table 1, the observed and calculated toxicities for the two phenols are quite close. The toxicity of toluene is much less than that predicted from equation (3). However, others give toxicities for toluene close to the calculated value of 0.097 mol m^{-3}. Buccafusco et al. (1981) give 0.14 mol m^{-3} as the concentration of toluene required for the acute toxicity to Bluegill (Lepomis machochirus) after 96 h at 22°C. Korn et al. (1979) measured the lethal doses after 96 h at 12°C of toluene and naphthalene for salmon fry and shrimps. For the fry (Oncorhynchus gorbuscha) they give 0.088 and 0.0097 mol m^{-3} respectively. For the shrimp (Eualus sp.) they give 0.15 mol m^{-3} for toluene, and 0.0076 mol m^{-3} for naphthalene in the shrimp (Pandolus gonicirus). The value calculated for naphthalene from equation (3) is 0.014 mol m^{-3}.

Alexander et al. (1978) observed that concentrations less than those required to kill the fish gave rise to adverse effects - loss of equilibrium; melanization; narcosis and swollen and

Table 1. Lethal doses for Pimephales promelis. Measured data from Alexander et. al., (LC50, 96 h) (1978) and Mayes et. al., (1983).

Formula	Compound	Lethal Dose ($mol\ m^{-3}$)	
		Measured	Calculated from equation (3)
$CHCl_3$	Trichloromethane	0.42	0.75
CH_2Cl_2	Dichloromethane	2.27	2.08
C_2Cl_4	Tetrachloroethene	0.11	0.12
C_2HCl_3	Trichloroethene	0.31	0.33
$C_2H_3Cl_3$	1,1,1-Trichloroethane	0.40	0.23
$C_2H_6O_2$	1,2-Ethanediol	854	1.84 (463)
C_3H_5ClO	1-Chloro-2,3-epoxypropane	0.13	0.82
$C_4H_{11}NO_2$	2,2'-Dihydroxydiethylamine	3.9	0.032 (19.3)
$C_6H_4Cl_2$	1,4-Dichlorobenzene	0.057	0.041
C_6H_5Cl	Chlorobenzene	0.23	0.12
C_6H_5ClO	4-Chlorophenol	0.033	0.07
C_6H_6O	Phenol	0.34	0.20
C_7H_8	Toluene	0.67	0.097

hemorrhaging gills. They measured the concentrations required to produce an effect and found these were approximately half the median lethal concentrations i.e. $Log_{10}C_B = 1.8$. It has been suggested (McGowan, 1955) that human beings should not be exposed for any considerable time to concentrations of chemicals corresponding to $Log_{10}C_B = 1.6$. These results on fish suggest that this value is about correct but it is, if anything, on the high side.

Many of the compounds tested for toxicity to fish have been phenols. As mentioned above, there is uncertainty about the interactions of these. Also many of them will be ionized to some extent under the conditions used for the tests. The un-ionized phenol is much more biologically active than the ion derived from it and corrections would have to be applied for this. However a good number of the compounds for which toxic concentrations were given by Heitmuller et al. (1981) were not phenols. These tests were carried out in sea-water at 25-31°C with sheepshead minnows (Cyprinodon variecatus) and 96 h exposure. In Figure 1, $-Log_{10}C_t$ values for the compounds consisting of carbon, hydrogen and halogens are plotted against 37500 V_x. The slope of 37500 is used in place of 36000 [equation (3)] in order to account for the salting out by the salts in sea-water (McGowan, 1968). The points in the figure fall near the line drawn for

$$-Log_{10}C_t = 375000\ V_x - 2.5 \qquad (4)$$

The intercept of -2.5 for $-Log_{10}C_B$ suggests that the sheepshead minnows may be somewhat more resistant to physically toxic

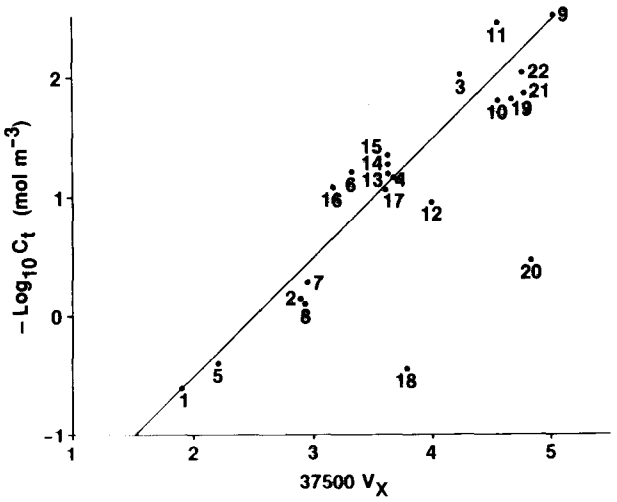


Figure 1. Toxicities (LC50, 96 h) to juvenile sheepshead minnows compared to values estimated from the molecular volume V_X. The straight line gives the values estimated from equation 3, with V_X calculated from atomic factors. The compounds are identified by number in Table 2.

compounds than the fathead minnows (see Table 1). The points (18) ethylbenzene and (20) 4-isopropyl-methylbenzene are so far from the line based on equation (4) that the tests on these should be re-examined.

The molecular volume V_X can be used as shown in Figure 1 to estimate the toxicities of compounds. Where the measured toxicity is less than the estimated toxicity, this may be due to interactions of polar groups with water for which appropriate corrections can be made using the interaction term E. Alternatively where the measured toxicity is much greater than the estimated toxicity, then the toxicity is due to specific chemical interactions rather than non-specific physical toxicity. The correlation can be used to predict the toxicity of new compounds and to design less toxic chemicals for industrial applications. This correlation may be useful in quantitative structure activity relationship (QSAR) models.

Physical toxicity depends on the physical properties of compounds and is non-selective. Selective toxicity may depend not only on factors such as the shape of the molecules and chemical reactivity of the compounds but on partitioning between phases as well (Blankley, 1983) and characteristic volumes may be useful in structure - activity relationships (QSAR) for such compounds.

Table 2. Toxicities (LC50, 96 h)to sheepshead minnows (Cyprinodon variegatus).[a]

Compound (Fig. 1)	Formula	$-\text{Log}_{10}C_t$	37500 V_x
1	CH_2Cl_2	-0.590	1.857
2	$CHBr_3$	0.149	2.907
3	C_2Cl_6	2.000	4.225
4	C_2HCl_5	1.244	3.766
5	$C_2H_2Cl_2$	-0.412	2.227
6	$C_2H_2Cl_4$	1.197	3.306
7	$C_2H_3Cl_3$	0.276	2.972
8	$C_3H_6Cl_2$	0.114	2.921
9	C_6HCl_5	2.495	5.002
10	$C_6H_2Cl_4$ (1,2,3,5)	1.770	4.543
11	$C_6H_2Cl_4$ (1,2,4,5)	2.432	4.543
12	$C_6H_3Cl_3$	0.936	4.083
13	$C_6H_4Cl_2$ (1,2)	1.180	3.625
14	$C_6H_4Cl_2$ (1,3)	1.276	3.625
15	$C_6H_4Cl_2$ (1,4)	1.301	3.625
16	C_6H_5Cl	1.051	3.166
17	C_8H_8	1.060	3.609
18	C_8H_{10}	-0.422	3.770
19	$C_{10}H_7Cl$	1.824	4.563
20	$C_{10}H_{14}$	0.444	4.833
21	$C_{10}H_{16}$	1.854	4.749
22	$C_{12}H_{10}$	1.854	5.018

[a] - The compounds are numbered as in Fig. 1. The measured toxicities ($-\text{Log}_{10}C_t$) are from Heitmuller et al. (1981).

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REFERENCES

Alexander HC, McCarty WM, Bartlett EA (1978) Toxicity of perchloroethylene, trichloroethylene, 1,1,1-trichloroethane and methylene chloride to fathead minnows. Bull Environ Contam Toxicol 20:344-352

Blankley CJ (1983) Introduction: A review of QSAR methodology. In: Topliss JG (ed) Quantitative structure-activity relationships of drugs. Academic Press, New York, p 1

Buccafusco RJ, Ellis SJ, Leblanc GA (1981) Acute toxicity of priority pollutants to bluegill (Lepomis macrochirus). Bull Environ Contam Toxicol 26:446-452

Ferguson J (1939) The use of chemical potentials as indices of toxicity. Proc Roy Soc 127:384-403

Ferguson J, Pirie H (1948) The toxicity of vapours to the grain weevil. Ann Appl Biol 35:532-550
Ferguson J (1964) The discovery of the anaesthetic halothane. Chemistry and Industry 818-824
Heitmuller PT, Hollister TA, Parrish PR (1981) Acute toxicity of 54 industrial chemicals to sheepshead minnows (Cyprinodon variegatus). Bull Environ Contam Toxicol 27:596-604
Korn S, Moles DA, Price SD (1979) Effects of temperature on the median tolerance limit of pink salmon and shrimp exposed to toluene, naphthalene and Cook Inlet crude oil. Bull Environ Contam Toxicol 21: 521-525
Mayes MA, Alexander HC, Dill DC (1983) A study to assess the influence of age on the response of fathead minnows in static acute toxicity tests. Bull Environ Contam Toxicol 31:139-147
McGowan JC (1951) The physical toxicity of chemicals. I. Vapours. J Appl Chem Lond 1:5120-5126
McGowan JC (1952) The physical toxicity of chemicals. III A systematic treatment of physical toxicity in aqueous solutions. J Appl Chem Lond 2:651-658
McGowan JC (1954) The physical toxicity of chemicals. IV Solubilities, partition coefficients and physical toxicities. J Appl Chem Lond 4:41-47
McGowan JC (1955) Physically toxic compounds and industrial hygiene. A M A Arch Ind Health II:315-323
McGowan JC (1968) Activity of physically toxic compounds in solutions. In Society of Chemical Industry Monograph No 29 Physico-chemical and biophysical factors affecting the activity of pesticides, p 141-158
McGowan JC (1978) Estimates of the properties of liquids. J Appl Chem Biotechnol 28:599-607
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