Bull. Environm. Contam. Toxicol. 23,579-586 (1979)

Rapid Nephelometric Estimate of Water Solubility of Highly Insoluble Organic Chemicals of Environmental Interest

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Environmental scientists have come to rely increasingly on physical and chemical properties of industrial chemicals (including pesticides) in attempts to predict their environmental behavior. However, the selection and determination of the appropriate properties are not always simple.

The recent work of CHIOU et al. (1977), LU and METCALF (1975), and METCALF et al. (1975) offers a simplified approach to predicting the tendency of organic chemicals to bioaccumulate based on the degree of water solubility. CHIOU et al. (1977) have developed an empirical equation for calculating *n*-octanol-water partition coefficients using aqueous solubilities. The formula was developed using a wide variety of chemicals, and is claimed to allow the prediction of partition coefficients to within an order of magnitude. The calculated partition coefficients can be used in predicting soil absorption (BRIGGS 1973), biological uptake (KENAGA 1972), lipophilic storage (DAVIES et al. 1975), and biomagnification (LU and METCALF 1975, METCALF et al. 1973, 1975; NEELY et al. 1974). CHIOU et al. have shown that for a number of chemicals, the water solubility is inversely proportional to bioconcentration in rainbow trout (*Salmo gairdneri*). This correlation has also been previously shown (LU and METCALF 1975, METCALF et al. 1975) for a number of other compounds using the mosquito fish (*Gambusia affinis*). Both of these studies reveal high biomagnification factors for the more insoluble organic substances, while polar, water-soluble, fat-insoluble compounds accumulate to a lesser extent, if at all.

The use of water solubility as a simple and rapid measure of an organic chemical's potential for bioaccumulating in the food chain has great appeal. However, numerous conflicting solubility values are given in the literature for many compounds of interest, and reliable water solubility data are lacking for many chemicals. One of the factors contributing to this situation is the lack of adequate methods for determining the water solubility of highly insoluble organic compounds. Many of the techniques typically used have limitations that restrict their widespread application. For example, radiotracer techniques require expensive tagged materials and sophisticated equipment. Spectrophotometric approaches are generally restricted to classes of compounds that can be measured at low levels by ultraviolet or fluorometric means, and are subject to interferences caused by the presence of minor constituents.

0007-4861/79/0023-0579 $01.60

One of the major sources of error in all of the above techniques is the preparation of the aqueous solution containing the solubilized test material. This is generally accomplished by exposure of an excess of the compound to water for an arbitrary period of time for equilibrium to be reached, followed by separation of the aqueous phase. Difficulties may be encountered due to several causes. For example, hydrolysis of the analyte may occur when long times are required for the substance and water to reach equilibrium. Another is the difficulty in obtaining the aqueous phase free of undissolved solute. Also, losses of analyte from the aqueous solution may occur because of adsorption onto filter media and containers.

This paper describes an approach that avoids many of the problems encountered in these other techniques. Based on earlier work by DAVIS and PARKE (1942) and BROOKER and ELLISON (1974), a nephelometric standard curve extrapolation technique is used to obtain water solubility data. The test substance is dissolved in a water-miscible solvent such as ethanol or acetone. Dilutions of increasing amounts of this solution with relatively large volumes of water give a series of turbid suspensions. The turbidity is measured nephelometrically and the relative intensity of scattered light is plotted vs. concentration of test material in each sample. Extrapolation of this standard curve to the relative intensity of a reagent blank gives an estimate of the experimental water solubility.

EXPERIMENTAL

Apparatus. Nephelometric measurements were made with a spectrophotofluorometer (Aminco-Bowman No. 4-8202) equipped with a photomultiplier photometer, a potted RCA IP28 photomultiplier tube, and a 150-W xenon arc lamp. All studies were performed at constant slit width, with the slit arrangement 3,2,2,3,5. Excitation and emission monochromators were adjusted to 600 nm in order to achieve maximum scattered light intensity. A standard 1-cm path length quartz fluorescence cuvette was used. Volumetric repipets and a syringe microburet (Micrometric Instrument Co., Model SB2) were used to prepare suspensions. A magnetic stirrer was used for mixing.

Reagents and Standard Solutions. Reagent grade ethanol and acetone were used as the solvents for all chemicals. Standard solutions were generally prepared by dissolving 0.25 g or less of test substance in 50 ml of absolute ethanol or acetone to give a concentration of 0.5% or less. This concentration was adjusted as necessary by serial dilution with the same solvent until between 0.5 and 1.5 ml of the standard solution produced a visibly turbid suspension in 40 ml of water. If the standard was too dilute, a more concentrated solution was prepared.

Distilled water (stored in closed repipet dispensers) was used for preparation of suspensions. Gum tragacanth (0.10 g) was dissolved in 1 L of hot distilled water, cooled, filtered, and stored in a closed repipet dispenser. The gum is an effective stabilizer for dispersions. Absence of dust or particles in all solutions was confirmed nephelometrically.

Procedure. Forty ml of water and 1 ml of gum solution were added to each of six to eight 50-ml beakers and mixed on a magnetic stirrer. A syringe microburet containing the test substance standard solution was positioned with the buret tip below the surface of a stirred water-gum solution. Test standard solution was added at the rate of about 0.1 ml/min until the first signs of turbidity appeared. The volume of test solution added was recorded and the concentration of the turbid solution calculated. The above steps were repeated using increasing quantities (about 0.1-ml increments) until six to eight turbid solutions had been prepared. All solutions were then allowed to "age" for a minimum of 15 min without stirring. Blanks were prepared similarly but without addition of any of the test material. Each sample was resuspended by stirring and transferred to the cuvette; nephelometric readings were taken and plotted against the sample concentration to produce a standard curve. Extrapolation of this curve to the blank reading yielded the solubility of the test compound. All determinations were conducted at room temperature (about 24° ± 2°C).

RESULTS AND DISCUSSION

Using the procedure described here, experimental water solubilities have been estimated for a number of chemicals of environmental interest. Practical grade compounds and mixtures, as well as pure chemicals, have been examined. These included representative hydrocarbons, phosphates, phthalates, chlorocarbons, and Aroclors. The experimentally determined solubilities of 48 chemicals are tabulated in Table 1 along with literature values for comparison. Our results represent the average of multiple determinations.

Accuracy and Precision. This procedure does have inherent limitations due to solvent, vapor pressure, and surface effects on formation of stable, turbid solutions. Its accuracy, however, is difficult to assess without absolute water solubility values. There are few available data on sparingly soluble organic compounds for comparison, and the validity of some of these is in question. We compared some of our experimental values with literature values obtained using similar approaches and sensitivities. For example, the experimental hydrocarbon data agree with those reported by DAVIS et al. (1942) who used a similar nephelometric technique. Similarly, there is agreement within an order of magnitude with literature values (LU et al. 1975, McKAY and LEINONEN 1975, METCALF 1976) based on radiotracer trechniques for such compounds as p,p'-DDT, tris(2,3-dibromopropyl) phosphate, and tri-p-cresyl phosphate. Based on these comparisons, we feel that this method may yield relatively good estimates of solubility for pure chemicals.

TABLE 1

Water Solubility of Selected Chemicals of Environmental Interest

Chemical	Purity	Water solubility, µg/L		Lit. Ref.
		Experimental	Literature	
Aromatic Hydrocarbons				
9,10-Dimethyl[α]benzanthracene	99%	55	45	a
9-Methyl[α]benzanthracene	99%	37	66	a
10-Methyl[α]benzanthracene	99%	11	55	a
Benz[α]anthracene	Practical	44	11	a
Pyrene	Practical	32	165	a
Chrysene	Practical	17	15	a
Fluoranthrene	99%	120	240	a
Biphenyl	99%	8,500	7,500	b
Diphenylmethane	Practical	3,000		
Chlorinated Hydrocarbons				
p,*p*'-DDT	99%	4	1-5, 1.2, 0.17	b-f
p,*p*'-DDD, olefin	99%	4		
p,*p*'-DDE	99%	65	40, 1.3	c,d
p,*p*'-TDE	99%	160		
p,*p*'-Kelthane	99%	1,200		
Lindane	99%	17,000	7,300, 10,000	b
Kepone	99%	7,600		
Methoxychlor	99%	40	100	g
Mirex	Practical	200		
Hexachlorobenzene	99%	110	6	d
Perthane	99%	100		
Chlorinated Hydrocarbon Mixtures				
Aroclor 1016	Multicomponent	49	490	h
Aroclor 1221	Multicomponent	590		
Aroclor 1242	Multicomponent	100	340, 240	b,h
Aroclor 1248	Multicomponent	60	54	b
Aroclor 1254	Mutlicomponent	57	12, 56	b,i

Aroclor 1260	Multicomponent	80	2.7	b
Aroclor 1262	Multicomponent	52		
Aroclor 1268	Multicomponent	300		
Aroclor 5432	Multicomponent	60		
Phthalates				
Diisodecyl phthalate	Multicomponent	280		
Didodecyl phthalate	Multicomponent	140		
Di-*n*-decyl phthalate	Practical	330		
Benzylbutyl phthalate	Practical	710		
Ditridecyl phthalate	Multicomponent	340		
Di(2-ethylhexyl) phthalate	Technical	285	100,000	j
Di(2-ethylhexyl) isophthalate	Practical	11		
Diphenyl phthalate	Practical	82		
Diisobutyl phthalate	Practical	6,200		
Trimethylcyclohexyl phthalate	Practical	120		
Dicyclohexyl phthalate	Practical	4,000		
Phosphates				
Diphenylmethyl phosphate	Practical	960		
Triphenyl phosphate	Practical	730	20,000	k
Diphenyl phosphate	Practical	>270,000		
Tris(2-ethylhexyl) phosphate	Practical	600		
Octyldiphenyl phosphate	Practical	140		
Tris(1,3-dichloroisopropyl) phosphate	Practical	7,000	19,200	d
Tris(2,3-dibromopropyl) phosphate	Technical	8,000		d
Tri-*p*-cresyl phosphate	Practical	74	260	

[a] DAVIS et al. 1942. [b] McKAY and LEINONEN 1975. [c] CHIOU et al. 1977. [d] METCALF 1976. [e] BOWMAN et al. 1960. [f] BIGGER et al. 1967. [g] RICHARDSON 1960. [h] EPA 1976. [i] HAQUE et al. 1974. [j] METCALF and LU 1973. [k] FONDYCE 1940.

On the other hand, the method yields values of uncertain accuracy for impure chemicals or mixtures of isomers and/or similar substances. The experimental values in Table 1 for Aroclors are in general agreement within an order of magnitude with published values (EPA 1976, HAQUE et al. 1974, McKAY and LEINONEN 1975). All have very low solubilities. Except for Aroclor 1268, whose measured solubility appears to be too great, the Aroclors exhibited decreasing solubility with increased chlorination. However, significant changes in solubility are not apparent with small differences in chlorine content (10-20%). Several industrial phthalate mixtures also showed decreasing solubility with increased molecular weight.

Based on these observations, it appears that this procedure gives experimental solubility values for mixtures that may be only approximations. The tendency to show some correlations with groups of closely related mixtures is encouraging, however. If the nature of the test samples is kept in mind, such approximations may still be useful.

In general, good precision (± 5%) was obtained for pure compounds in replicate measurements. Results were less precise for the Aroclors and practical grade materials (about ± 20%). Precision was poor for compounds such as lindane and hexachlorobenzene. In each case, excessive particle growth occurred. In addition, scum formation occurred over the lindane suspensions. These aberrations probably contributed to the poor results. Volatile liquids such as benzene, toluene, and xylene gave extremely poor precision, possibly because they escape too rapidly from stirred water solutions. The procedure appears unsuitable for measuring the water solubility at room temperature of volatile materials such as these.

Application Criteria. A given test material must meet two principal criteria for this procedure to be suitable. First, the particles of the substance must differ from water in refractive index so that light-scattering readings can be made. Second, the substance should form dispersions which are stable for a reasonable time (30-60 min) so that reliable nephelometric measurements may be taken. Slow rates of precipitation, crystal growth, and particle coagulation contribute to the formation of more stable dispersions. Colligative property effects have been discussed in greater detail by DAVIS and PARKE (1942).

Experimental Parameters. Good precision is dependent on proper aging of the dispersion solution at a reasonably constant temperature. Selected dispersions were aged 0, 15, 30, and 60 min at room temperature (about 24° ± 2°C). Scattered light intensity for the test substances peaked after about 15 min and remained constant up to about 30 min, after which it began to decrease. As aging increases, an equilibrium between solution and suspended particles is approached. Following this state, crystal growth and coagulation become more pronounced. The use of gum tragacanth slows this latter process. Based on our observations, we concluded that 15-min aging was adequate for most of the substances encountered in this study. All compounds will not approach equilibrium at the same rates, and ideally should be checked individually to determine optimum aging requirements.

The effect of ethanol and acetone on solubility measurements appeared negligible up to concentrations of about 5%. Higher levels were not tested. These solutions also gave a horizontal nephelometric baseline equivalent to water-gum solutions alone, i.e., the presence of solvent did not affect background light scatter. As a result, water-gum solutions were routinely used as blanks.

The method described here provides a rapid and simple means for estimating the water solubilities of pure chemicals and the "apparent" solubility behavior of less pure compounds and mixtures. It allows rapid comparisons of a test material with similar chemicals that are known environmental hazards. It permits quick classification of a substance as to its relative solubility range which can then used in the empirical equations of CHIOU et al. (1977) and NEELY et al. (1974) to predict octanol-water partitionm behavior and bioaccumulation factors. In such cases, the uncertainties in the empirical equations are so great compared to that of the solubility value that an order of magnitude variance in solubility has only a small impact on the significance of the result. Consequently, the approximate solubilities given by this method for most chemical mixtures as well as pure chemicals may be sufficiently accurate for many purposes.

This procedure offers an alternative approach to slower, more expensive, and sophisticated methods of determining water solubilities of relatively insoluble pure compounds. It also provides a means of obtaining rough solubility estimates of impure chemicals and mixtures. It is thought to be relatively accurate for pure chemicals, and fairly reliable as an indicator of the approximate solubility behavior of many mixtures, but not suitable for volatile or relatively soluble materials. The data obtained may be useful for predicting octanol-water partition behavior, which in turn may serve to assess a chemical's potential for environmental biomagnification.

ACKNOWLEDGMENT

The author wishes to thank Ivan Egry for technical assistance and Pasquale Lombardo and Sidney Williams for helpful advice and editorial comment.

REFERENCES

BIGGER, J.W., G.R. DUTT, and R.L. RIGGS: Bull. Environ. Contam. Toxicol. 2, 90 (1967).

BOWMAN, M.C., F. ACREE, JR., and M.K. CORBETT: J. Agric. Food Chem. 8 (5), 406 (1960).

BRIGGS, G.G.: "A Simple Relationship Between Soil Absorption of Organic Chemicals and Their Octanol/Water Partition Coefficients," Proceedings of the 7th British Insecticide and Fungicide Conference, 1973.

BROOKER, P.J., and M. ELLISON: Chem. Ind. (London) 19, 785 (1974).

CHIOU, C.T., V.H. FREED, D.W. SCHMEDDING, and R.L. KOHNERT: Environ. Sci. Technol. 11 (5), 475 (1977).

DAVIES, J.E., A. BARQUET, V.H. FREED, R. HAQUE, C. MORGADE, R.E. SONNEBORN, and C. VACLAVEK: Arch. Environ. Health 30, 608 (1975).

DAVIS, W.W., and T.V. PARKE, Jr.: J. Am. Chem. Soc. 64, 101 (1942).

DAVIS, W.W., M.E. KRAHL, and G.H.A. CLOWES: J. Am. Chem. Soc. 64, 108 (1942).

Environmental Protection Agency, Environmental Research Laboratory, Athens, Ga., Quarterly Summary, No. 8, July-Sept. 1976.

FONDYCE, G.R.: Ind. Eng. Chem. 32, 1054 (1940).

HAQUE, R., D.W. SCHMEDDING, and V.H. FREED: Environ. Sci. Technol. 8 (2) 139 (1974).

KENAGA, E.E.: Residue rev. 44, 73 (1972).

LU, P.Y. and R.L. METCALF: Environ. Health Perspect. 10, 269 (1975).

LU, P.Y., R.L. METCALF, A.S. HIRWE, and J.W. WILLIAMS: J. Agric. Food Chem. 23 (5), 967 (1975).

McKAY, D. and P.J. LEINONEN: Environ. Sci. Technol. 9 (13), 1178 (1975).

METCALF, R.L.: "Evaluation of the Utility of the Model Ecosystem for Determining the Ecological Fate of Substances Subject to FDA Regulatory Authority," Final Report, FDA Contract 74-127, Dec. 1976.

METCALF, R.L. and P.Y. LU: "Environmental Distribution and Metabolic Fate of Key Industrial Pollutants and Pesticides in a Model Ecosystem," Research Report No. 69, University of Illinois Water Resources Center, 72 (1973).

METCALF, R.L., I.P. KAPOOR, P.Y. LU, C.K. SCHUTH, and P. SHERMAN: Environ. Health Perspect. 4, 35 (1973).

METCALF, R.L., J.F. SANBORN, P.Y. LU, and D. NYE: Arch. Environ. Contam. Toxicol. 3, 151 (1975).

NEELY, W., D.R. BRANSON, and G.E. BLAU: Environ. Sci. Technol. 8 (13), 1113 (1974).

RICHARDSON, L.T.: Can. J. Bot. 38, 163 (1960).