

Mobilization of Azaarenes from Wastewater Treatment Plant Biosludge

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Azaarenes are a class of polycyclic aromatic bases commonly found in fossil fuel combustion residues (Sawicki et al. 1965) and products and residues from direct coal liquefaction processes (Kimball and Munro 1981). Many azaarenes are carcinogenic (Searle 1976), and they also exhibit mutagenic activity in microbial mutagenesis bioassays (Ho et al. 1981). Much of the mutagenic activity of coal derived petroleum substitutes is found in the isolate fraction containing azaarenes and polycyclic primary amines (Guerin et al. 1980, 1981).

Biological sludges produced by wastewater treatment plants receiving inputs containing azaarenes are likely to contain these compounds, due to their biological accumulation potential (Southworth et al. 1980). Extracts of such sludge have revealed the presence of quinoline (Maskarinec et al. 1982), and other azaarenes are undoubtedly present. Biosludges produced from treating wastewaters of the SRC-II coal liquefaction process proved to be mutagenic in microbial bioassays (Keleti et al. 1982).


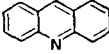
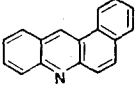
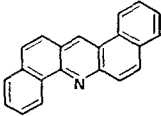
The possible presence of azaarenes in residues disposed of in landfills represents a potential source of groundwater contamination. Assessment of the hazard posed by azaarenes in such wastes requires a knowledge of the ease with which such materials are leached from the residue by infiltrating water. Since the partitioning of aromatic bases between a sorbent and water would be expected to vary with pH, it is necessary to know the partitioning behavior of both the neutral bases and their conjugate acids. The objective of this study was to investigate the partitioning behavior of a homologous series of azaarenes (Table 1) between water and a wastewater sludge under conditions representing both neutral and cationic species of the aromatic bases.

MATERIALS AND METHODS

The studies were conducted using four azaarenes representing a wide range in anticipated partitioning behavior (Southworth et al. 1980). Radioisotopically labelled quinoline (phenyl

^{14}C , 1.53×10^8 Bq/mmole), acridine (phenyl- ^{14}C , 1.53×10^8 Bq/mmole), benz(a)acridine (7^a, 8-, 9-, 10-, 11-, 11⁹- ^{14}C , 1.75×10^8 Bq/mmole) and dibenz(a,h)acridine (1,2 benzo- $^{14}\text{C}(u)$, 3.01×10^8 Bq/mmole) obtained from New England Nuclear Corp. were utilized in the studies. The biosludge was a stabilized biosludge filter cake produced at the H-Coal pilot plant in Cattlettsburg, Kentucky (Griest et al. 1980). The sludge was highly alkaline, containing 3.5 meq/g dry weight alkalinity, and contained Ca^{+2} and Fe^{+3} as major cations. Organic carbon content was approximately 14%, oil content was 4%, and water content was ~60%. While such a material cannot be assumed to be representative of residues produced by any other facility, its constituents (microbial biomass, oil, and inorganic flocculants) would be typical of most such residues, although relative proportions would certainly vary.

Table 1. Properties of Test Compounds

	Structure	K_{ow}^a	pK_a^b
Quinoline		98	4.5
Acridine		1,995	5.6
Benz(a)acridine		28,184	5.2
Dibenz(a,h)acridine		398,107	3.6

^aOctanol-water partition coefficient, calculated from Leo, 1975.

^bIonization constant of conjugate acid.

Partitioning measurements were made by adding known amounts of the test compounds to suspensions of the filter cake in distilled water. Stock solutions of the test compounds were made up in methanol and added to the suspensions with the concentration of methanol in water never exceeding 500 mg/l. The suspensions were shaken on a wrist action shaker for two hours and then centrifuged at 400 g for 10 minutes. Preliminary experiments indicated that sorptive equilibrium was attained in less than twenty minutes. A one or two ml aliquot of supernatant was removed from each sample and counted on a liquid scintillation spectrometer (Packard Tri-carb Model 4640). The extent of

sorption was determined by comparing ^{14}C remaining in solution in centrifuged suspensions with controls containing the same initial concentration of test compound but no suspended solids. The solid-water ratio was varied from compound to compound to optimize accuracy of the sorption measurements by having approximately a 50% decrease in solute concentration. Suspended solids concentrations ranged from 50 g/l to 0.1 g/l.

Sorption of the neutral species was carried out by adding sludge to distilled water, with no adjustment to pH. A range of pH from 10.3-12.0 was observed depending on solid:water ratio. This range was well above the pK_a of the conjugate acids of these compounds. Sorption of the conjugate acids was investigated at pH 3.0 by adjusting the pH of the suspensions with 1 N HCl. Quinoline, acridine, and benz(a)acridine, with pK_a 's > 4.5 were predominantly cationic at this pH, while dibenz(a,h)acridine (pK_a estimated at 3.6 by fluorimetric procedure) was still partially neutral. Therefore, the partitioning of the conjugate acid of dibenz(a,h)acridine was repeated at pH 2.

Partitioning measurements of the neutral compounds were made over a range of concentrations to verify that K_p was independent of concentration. Since this was observed to be the case, a single concentration was used to measure K_p at pH 3. Concentration ranges were: quinoline (3.5 - 13.7 $\mu\text{g/l}$), acridine (2.6 - 35.0 $\mu\text{g/l}$), benz(a)acridine (4.3 - 17.3 $\mu\text{g/l}$) and dibenz(a,h)acridine (6.0 - 21.2 $\mu\text{g/l}$). Concentrations employed for cationic species were: quinoline (86 $\mu\text{g/l}$), acridine (18.1 $\mu\text{g/l}$), benz(a)acridine (135 $\mu\text{g/l}$) and dibenz(a,h)acridine (14.0 $\mu\text{g/l}$).

The partitioning of organics between water and autoclaved yeast has been used as a model for sorption by microbial biomass or detritus (Herbes, 1977). The sorption of the four test compounds by autoclaved yeast was investigated using the methods and solid:water ratios employed for the biosludge, in an attempt to describe hydrophobic sorption on microbial biomass versus azaarene chemical structure.

A preliminary study to ascertain the possible importance of cation exchange processes in the sorption of acridine by soil was carried out using ^{14}C labelled acridine and a reference clay mineral (Montmorillonite 26, Ward's, Rochester, New York). Acridine was added to 0.5 g/l clay suspensions buffered with .001 M sodium acetate - bicarbonate to pH 5, 6, and 7.5.

The concentration of Ca^{+2} was adjusted to 1.0, 0.1, or .01 M by adding CaCl_2 from a stock solution. Samples were maintained 20 hours on a wrist action shaker at 25°C and processed thereafter the same as the sludge samples. Initial acridine concentration was 0.6 mg/l, duplicate determinations were made at each combination of pH and (Ca^{+2}).

RESULTS AND DISCUSSION

Large differences in sorption were observed among the four compounds (Table 2). Quinoline, the least hydrophobic of the compounds, was sorbed the least by the sludge, with $K_p = 31.5$ and 1.5 for the neutral and cationic species, respectively. The addition of one aromatic ring to a previous structure seems to increase partitioning by roughly a factor of ten. Thus, for neutral azaarene, K_p goes from 31.5 for quinoline (two rings) to 433 for acridine (three rings), 7,067 for benz(a)acridine (4 rings) and 55,167 for dibenz(a,h)acridine. A similar pattern is seen for the cationic species (pH 3), with the exception of dibenz(a,h)acridine, which was not fully protonated at pH 3.

Table 2. Partitioning of azaarenes between water and biosludge. K_p for neutral (pH > 10) and cationic species (pH < 3.0) \pm standard deviation of estimate (dry weight basis)

	Neutral	Cationic
Quinoline (1:100, 1:20)*	31.5 \pm 2.6	1.47 \pm 1.09
Acridine (1:1000, 1:100)	433 \pm 113	11.2 \pm 4.7
Benz(a)acridine (1:1000, 1:1000)	7,067 \pm 859	184 \pm 43
Dibenz(a,h)acridine (1:10,000, 1:1000)	55,167 \pm 3856	2059 \pm 258**

*Solid:water ratio (wet weight) of experimental determination. Replicates - Neutral, n = 8, cationic, n = 3.

**pH 2, 6665 \pm 279 at pH 3.

The cationic species were much less readily sorbed than the neutral azaarenes. Partition coefficients (K_p) for the protonated compounds were generally about a factor of 30 less than what was observed at high pH (Table 2). Thus, acidic conditions would be an important factor in increasing the mobilization of these aromatic bases from disposal sites. The low K_p 's of quinoline and acridine (1.5 and 11.2, respectively) suggest that they would be readily leached under acid conditions, while benz(a)acridine would be substantially mobile.

The partitioning of azaarenes between organic rich sludge and water appears to be dominated by the mechanism of hydrophobic partitioning. Thus, K_p for these compounds vs the biosludge varies with chemical structure in the same way as other measures

of partitioning between water and other phases. In Figure 1, K_p (neutral azaarenes) is plotted as the independent variable against other measures of partitioning for the same compounds such as octanol-water partition coefficient (K_{ow}), bioconcentration factor (Daphnia), partition coefficient vs autoclaved yeast, and K_p (cationic azaarenes). A very strong relationship is seen between K_p (neutral azaarenes) and the other partitioning measures. Such correlation is not unexpected if hydrophobic partitioning is the dominant mechanism determining sorption (Kenaga, 1979; Mackay, 1982).

Low molecular weight azaarenes (2-3 rings) are readily leached from organic rich sludge, while under acidic conditions the mobilization of larger compounds becomes more significant. The large amount of lime and ferric chloride used to stabilize and flocculate the sludge would ensure the presence of alkaline conditions initially upon disposal. Leaching of the inorganic bases and generation of CO_2 and organic acids as the sludge degrades could act to promote the eventual establishment of a low pH environment within a disposal site.

The high correlation between K_p and other fugacity based partition measures indicates that predictions of partition coefficients for other azaarenes and organic rich wastes should be possible based on knowledge of k_{ow} , pK_a , and biomass or carbon content of the waste. It is not the purpose of this study, however, to develop such a predictive capability, and extrapolation of these data to other compounds and wastes should be viewed as a rough first approximation.

The mobility of azaarenes outside the disposal site will be determined in part by the sorption of the compounds by soil receiving the leachate from a disposal site. Predicting such sorption from a knowledge of soil organic carbon content and k_{ow} is possible if hydrophobic partitioning is the mechanism of sorption (KARICKHOFF et al. 1979). However, studies of the sorption of acridine by a number of soils (BANWART et al. 1982) indicated that such soil-carbon based predictions underestimated the sorption of acridine by about a factor of ten. When the sorption of acridine by a reference clay mineral was investigated, it appeared to be readily sorbed by a cation exchange mechanism (Figure 2). Sorption was highest under conditions of high acidity and low ionic strength, and decreased at higher pH and higher ionic strength. Thus, the conditions which maximize mobilization of acridine from organic rich wastes would act to immobilize it in clay soils.

The data presented in this paper are only a part of the information needed to assess the hazard to groundwater of azaarenes in organic rich sludges disposed of in landfills. Information on azaarene concentrations in the waste, amount of waste disposed, water infiltration rates, leachate dilution and dispersion, rates of microbial degradation, and attenuation by soil adsorption are necessary information needed to predict the

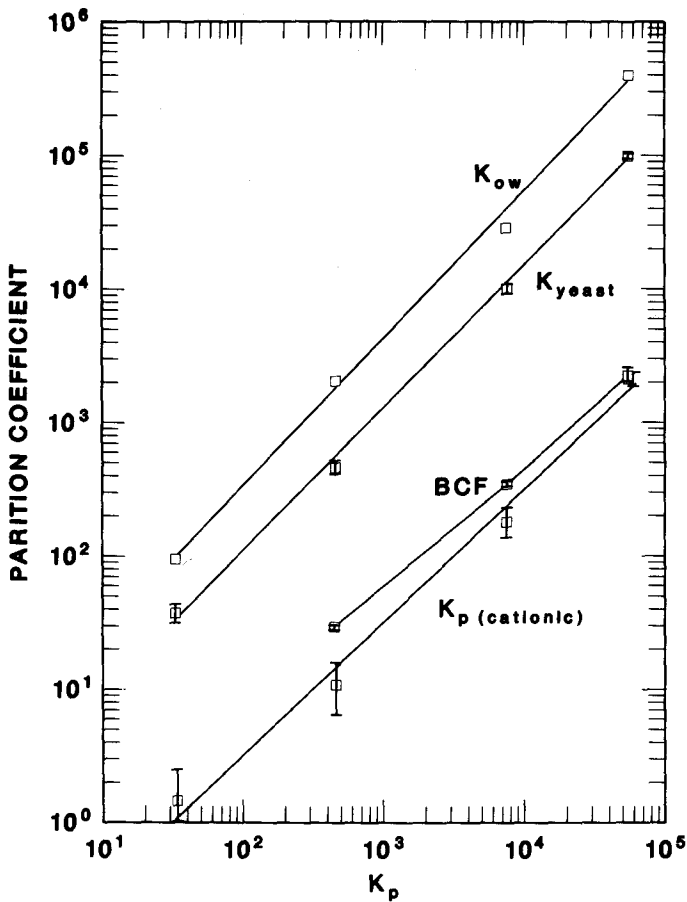


Fig. 1. Partition coefficients describing octanol-water partitioning (K_{ow}), autoclaved yeast-water partitioning (K_{yeast}), *Daphnia pulex* bioconcentration (BCF), and biosludge-water partitioning of protonated azaarenes [K_p (cationic)] vs biosludge-water partition coefficients for the neutral azaarenes [quinoline, acridine, benz(a)acridine, and dibenz(a,h)acridine].

potential for groundwater contamination. Much of this information remains to be obtained.

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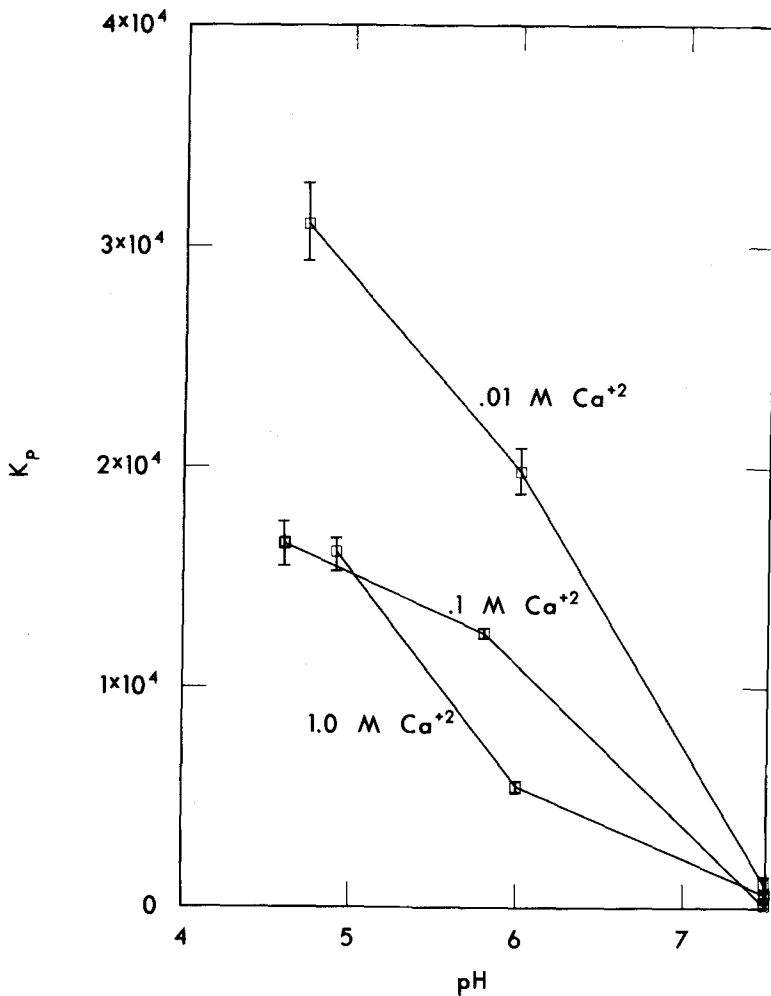


Fig. 2. Influence of pH and $[Ca^{+2}]$ on the partition coefficient (K_p) describing the sorption of acridine from water by Wyoming bentonite (montmorillonite #26, 0.5 g/l).

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