# A Microscopic Hydrophobicity Parameter

**F. M. Menger* and U. V. Venkataram**

*Contribution from the Department of Chemistry, Emory University, Atlanta, Georgia 30322.*
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**Abstract:** *p*-Nitrophenyl laurate at $1 \times 10^{-5}$ M in water forms aggregates within which the ester groups hydrolyze slowly (about $10^3$ less than a short-chain monomer). Salts of the general structure $RNMe_3^+X^-$ disrupt or destroy the aggregates; the ester groups are thereby "deshielded", and the observed hydrolysis rate increases. The magnitude of the rate increase at a given salt concentration depends on R: the more hydrophobic the R group, the greater the rate enhancement. This observation provided the basis of a "microscopic" hydrophobicity parameter MH which was evaluated for 25 different Rs (e.g., MH = 0.73, 0.97, and 1.33 for R = ethyl, *n*-butyl, and *n*-hexyl). MH values were used to assess the role of branching, unsaturation, cyclization, aromaticity, halogenation, etc., in hydrophobic association. The parameters correlate well with Hansch $\pi$ values for aliphatic substituents but not for aromatic groups. Since the MH scale is based on the specific binding of one molecule to another, it may be well suited for modeling association among bioactive species.

Dielectric constant and $E_T(30)$ (two measures of solvent polarity) illustrate the distinction between macroscopic and microscopic parameters. The dielectric constant is obtained from the change in capacitance upon immersing metal plates into a liquid; $E_T(30)$ is based on the solvent-dependent absorption of a dye.[1] Since dielectric constant reflects solvent behavior in the bulk, it is termed a macroscopic parameter. On the other hand, $E_T(30)$ represents solvent interactions at the molecular level and hence is called a microscopic constant. Not surprisingly, dielectric constant and $E_T(30)$ correlate poorly. In general, $E_T(30)$ is far more useful than dielectric constant for treating solvent effects on rates, equilibria, and spectra.

The ensuing article describes a microscopic hydrophobicity scale for 25 organic substituents. Although hydrophobicity scales have already been developed,[2,3] they rely on partitioning phenomena and are thus macroscopic in character. Consider the important Hansch hydrophobibity parameter $\pi_X$ defined in eq 1.[4,5] $P_X$ and $P_H$ are the partition coefficients between 1-octanol and water for

$$\pi_X = \log P_X - \log P_H \quad (1)$$

$C_6H_5X$ and $C_6H_6$, respectively. A positive $\pi_X$ (such as +0.56 for $CH_3$) means that the substituent favors the organic phase. If X has a negative $\pi_X$ (such as −0.67 for OH), then the substituent causes partitioning into water relative to a hydrogen. Hansch $\pi_X$ parameters have been widely applied to quantitative structure/activity relationships in pharmacology. Success has been achieved primarily when bioactive molecules are "nonspecific", i.e., when transport dominates their function. This seems to be the case for barbituate action where drug potency and lipophilicity correlate significantly.[6] Of course, if physiological action depends on specific interactions at an active site or at a receptor site, then activity vs. $\pi_X$ correlations fail; stereoelectronic terms must be introduced additionally to secure meaningful structure/activity relationships.[7]

Another problem with $\pi_X$ relates to the complexity of the distribution mechanism which, according to Hansch and Leo, is governed by hydrogen bonding forces and by solute molecular volume.[8] Thus, $\pi_X$ can be used only with closely related congeners. With structurally diverse biomolecules, the Hansch approach often breaks down. Owing to these considerations, we felt it worthwhile to devise a hydrophobicity scale with a totally different origin. As will be described, this scale is based on the hydrophobic association of additives with a long-chain ester. Since the resulting hydrophobicity parameters arise from specific intermolecular attraction, they are microscopic in nature and not necessarily correlatable with $\pi_X$. Such a new scale could be useful in pharmacology,[9] toxicology,[10] protein structure,[11] membrane behavior,[12] and polymer chemistry[13] —in short wherever hydrophobicity plays a role.

In 1968 we observed[14] that the rate constants for basic hydrolysis of *p*-nitrophenyl laurate (pNPL) decrease with increasing initial ester concentration in the range of $10^{-6}$ to $10^{-5}$ M. Evidently, the ester molecules bind hydrophobically to one another, thereby enclosing the ester head groups among hydrocarbon chains where hydrolysis is retarded. Rate inhibitions are appreciable: the second-order rate constant for basic hydrolysis of pNPL at $1.0 \times 10^{-5}$ M is almost three orders of magnitude smaller than that for *p*-nitrophenyl acetate! We also observed, and this is important for the present study, that additives (e.g., urea and dioxane) in the water can accelerate the pNPL hydrolysis. For example, 8.0 M urea increases the rate of pNPL hydrolysis 33-fold (while actually inhibiting *p*-nitrophenyl acetate hydrolysis). The additive-induced accelerations are best explained by perturbation or destruction of the aggregates (Figure 1) which exposes the ester groups to hydroxide attack.

"Unmasking" the ester moiety by an additive should depend on how effectively the additive interacts with the pNPL chains. This in turn depends on the hydrophobicity of the additive. In other words, an additive with a butyl group should enhance the ester hydrolysis to a greater extent than the corresponding additive bearing only a methyl. It was immediately clear that if we were to examine a wide variety of groupings (including ones such as cyclohexyl and benzyl), we would need to attach them to a water-solubilizing entity. A quanternary nitrogen was selected for this purpose. Thus, our additives all consisted of $RNMe_3^+X^-$ where R is a collection of 25 different groups (Table I). In line with expectations, the more hydrophobic groups produced the larger rate increases, and the foundation for a kinetically based hydrophobicity parameter was therefore in hand.

## Experimental Section

Trimethylamine (25% solution in methanol) was obtained from Matheson Coleman & Bell. All other compounds required for the syn-

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**Table I.** Preparation Method, Melting Point, and Elemental Analysis for Quaternary Ammonium Salts $RN^+(Me_3)X^-$

| | | | | | analysis | | | | |
|---|---|---|---|---|---|---|---|---|---|
| no. | R | X | method[a] | mp, °C | C | H | N | X | ref |
| **1** | methyl | Br | *b* | | | | | | |
| **2** | ethyl | I | A | >300[c] | 27.90 | 6.56 | 6.49 | *e* | *g* |
| | | | | >300[d] | 27.92 | 6.56 | 6.51 | *f* | |
| **3** | *n*-propyl | I | A | 190 | 31.51 | 7.05 | 6.09 | | *g* |
| | | | | 189 | 31.46 | 7.04 | 6.11 | | |
| **4** | isopropyl | I | A | >300 | 31.50 | 7.08 | 6.09 | | *h* |
| | | | | 316 | 31.46 | 7.04 | 6.11 | | |
| **5** | *n*-butyl | I | A | 231 | 34.69 | 7.48 | 5.72 | | *g* |
| | | | | 226 | 34.58 | 7.46 | 5.76 | | |
| **6** | *tert*-butyl | I | B | 256 | 34.66 | 7.47 | 5.72 | | *h* |
| | | | | 260 | 34.58 | 7.46 | 5.76 | | |
| **7** | *n*-pentyl | I | A | 224 | 37.28 | 7.86 | 5.41 | | |
| | | | | | 37.37 | 7.84 | 5.45 | | |
| **8** | 1-methylbutyl | I | A | 224 | 37.48 | 7.87 | 5.43 | 49.17 | |
| | | | | | 37.37 | 7.84 | 5.45 | 49.34 | |
| **9** | *n*-hexyl | I | A | 160 | 39.68 | 8.19 | 5.11 | | *g* |
| | | | | 166 | 39.86 | 8.18 | 5.16 | | |
| **10** | cyclopentyl | I | C | 271 | 37.78 | 7.13 | 5.41 | 49.63 | |
| | | | | | 37.66 | 7.11 | 5.47 | 49.74 | |
| **11** | cyclohexyl | I | B | 271 | 40.25 | 7.48 | 5.18 | | |
| | | | | | 40.16 | 7.49 | 5.20 | | |
| **12** | allyl | I | A | 104 | 31.81 | 6.22 | 6.15 | 55.76 | *g* |
| | | | | 102 | 31.73 | 6.21 | 6.17 | 55.89 | |
| **13** | 2-propynyl | I | B | 181 | 32.09 | 5.40 | 6.19 | 56.29 | |
| | | | | | 32.01 | 5.37 | 6.22 | 56.39 | |
| **14** | 3-butenyl | I | A, D | 236 | 34.93 | 6.70 | 5.78 | 52.56 | |
| | | | | | 34.87 | 6.69 | 5.81 | 52.63 | |
| **15** | phenyl | I | *b* | 227 | 41.12 | 5.38 | 5.31 | 48.15 | |
| | | | | 227 | 41.08 | 5.36 | 5.32 | 48.23 | |
| **16** | benzyl | I | B | 179 | 43.33 | 5.82 | 5.00 | | *f* |
| | | | | 179 | 43.33 | 5.82 | 5.05 | | |
| **17** | 4-chlorobutyl | I | A, D | 182 | 29.42 | 5.97 | 5.01 | | |
| | | | | | 30.28 | 6.17 | 5.05 | | |
| **18** | 4-bromobutyl | I | A, D | 131 | 25.30 | 5.40 | 4.62 | | |
| | | | | | 26.10 | 5.32 | 4.35 | | |
| **19** | 4-iodobutyl | I | A | 160 | 23.43 | 4.75 | 4.00 | 67.80 | |
| | | | | | 22.78 | 4.64 | 3.79 | 68.79 | |
| **20** | 2-ethoxyethyl | Br | A | 174 | 39.07 | 8.44 | 6.49 | 38.48 | |
| | | | | | 39.63 | 8.55 | 6.60 | 37.67 | |
| **21** | 2-phenoxyethyl | Br | A | 162 | 50.74 | 6.98 | 5.34 | 30.79 | |
| | | | | | 50.78 | 6.97 | 5.38 | 30.71 | |
| **22** | *p*-methylbenzyl | Br | A | 197 | 53.97 | 7.78 | 5.66 | 32.49 | |
| | | | | | 54.10 | 7.43 | 5.74 | 32.72 | |
| **23** | *p*-fluorobenzyl | Br | A | 237 | 48.32 | 6.10 | 5.61 | | |
| | | | | | 48.40 | 6.09 | 5.65 | | |
| **24** | *p*-chlorobenzyl | Br | A | 207 | 45.39 | 5.71 | 5.29 | | |
| | | | | | 45.39 | 5.75 | 5.26 | | |
| **25** | *p*-bromobenzyl | Br | A | 220 | 38.93 | 4.92 | 4.52 | 51.59 | |
| | | | | | 38.86 | 4.89 | 4.53 | 51.71 | |

[a] See Experimental Section. [b] Purchased from Aldrich Chemical Co. [c] Experimental melting point (uncorrected). [d] Literature melting point. [e] Observed. [f] Calculated. [g] Kato, T.; Morikawa, T.; Suzuki, Y. *J. Pharm. Soc. Jpn.* **1952**, *72*, 1177. [h] Grovenstein, E., Jr.; Blanchard, E. P.; Gordon, A. D.; Stevenson, R. W. *J. Am. Chem. Soc.* **1959**, *81*, 4842. [i] Brasen, W. R.; Hauser, C. R. *Organic Syntheses*; Wiley: New York, 1963; Collect. Vol. IV, pp 585.

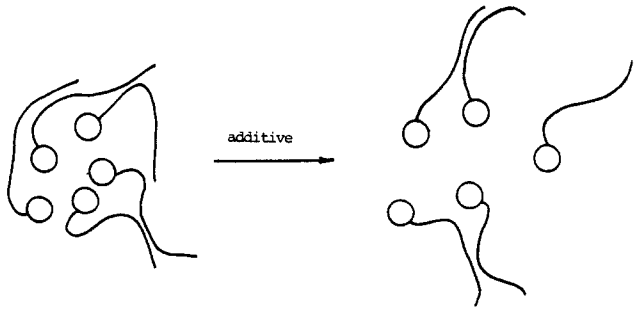



**Figure 1.** Schematic representation of a *p*-nitrophenyl laurate aggregate being disrupted by an additive in the aqueous solvent. The perturbation causes a marked increase in the ester hydrolysis rate.

thesis of the quaternary ammonium salts in Table I were purchased from Aldrich Chemical Co. and used without further purification. Sigma Chemical Co. supplied the *p*-nitrophenyl acetate and the *p*-nitrophenyl laurate (pNPL). Both gave a satisfactory yield of *p*-nitrophenolate, as judged spectrophotometrically, when hydrolyzed in base. Acetonitrile was distilled over calcium hydride. Microanalyses were performed by Atlantic Microlab, Atlanta, GA.

**Syntheses of Quaternary Ammonium Salts.** The $RNMe_3^+X^-$ salts in Table I were prepared according to one of the following procedures.

**Procedure A.** When an alkyl iodide (or bromide in the case of compounds **14**, **17**, **18**, and **20–25** in Table I) was commercially available, it was treated with an excess of trimethylamine (halide:amine = 1:1.3) for 24–48 h at room temperature. No solvent was used other than the methanol accompanying the trimethylamine. The reaction slurry was poured into five times the volume of ether and stirred. Solid product was removed by filtration and washed repeatedly with ether. Crystallization was carried out twice from ethanol–ether mixtures with activated charcoal during the first purification. Crystals (obtained in roughly 80% yield) were dried at 80 °C under 1 mm of pressure overnight.

**Procedure B.** When a tertiary amine $RNMe_2$ was available, the compound (10–25 g) was dissolved in 100 mL of absolute ethanol and cooled in an ice bath. A slight molar excess of methyl iodide was then added with stirring over a 30-min period after which the reaction was continued for 24–48 h at room temperature. Workup of the product was identical with that given in procedure A.

**Procedure C.** Cyclopentyltrimethylammonium iodide was prepared by refluxing cyclopentylamine (10 g) with methyl iodide (60 g) in absolute ethanol (100 mL) for 48 h. The reaction mixture was cooled, poured into an excess of ether (500 mL), and filtered; recrystallization of the solid from ethanol–ether gave the quaternary ammonium salt in modest yield.

**Procedure D.** Three quaternary ammonium bromides were converted into the corresponding iodides (compounds **14**, **17**, and **18**) by ion exchange chromatography. Amberlite IRA-400 (300 g, 4 mequiv/g capacity) in the chloride form was placed in a 45 × 5 cm column and treated with a liter of 20% aqueous KI at a slow flow rate. The resin was then washed with deionized water until free of iodide (silver nitrate test). A 10% aqueous solution of a quaternary ammonium bromide was introduced onto the column; subsequent elution with water at 1 mL/min gave the quaternary ammonium iodide which was recovered from the water with the aid of a rotary evaporator attached to a vacuum pump. Crystallization was effected with ethanol–water mixtures.

Table I lists the melting points and analytical data for the 25 salts.

**Kinetics.** The observed rate constant for hydrolysis of *p*-nitrophenyl laurate (pNPL) was obtained in the following manner. A 1.00-cm stoppered cuvette was filled with 3.00 mL of 0.100 N NaOH and equilibrated at 25.0 ± 0.1 °C in a Cary 14 spectrophotometer set at 400 nm (0.1–0.2 slidewire) for greater than 15 min. pNPL in acetonitrile (44 μL) was then introduced into the cuvette (by means of a small stirring rod flattened at one end) such as to give an initial ester concentration in the cuvette of $9.80 \times 10^{-6}$ M. The increase in absorbance was next traced as a function of time until completion of the reaction (>8 half-lives). First-order rate constants were obtained in the usual manner with a Hewlett-Packard 9830A calculator-plotter. Linearity in the plots was satisfactory ($r = 0.9990$) for the first half-life of the hydrolysis.

Hydrolysis rates of pNPL in the presence of quaternary ammonium salts were determined similarly. The main difference was that cuvettes were filled with 3.00 mL of a solution containing 0.1–0.6 M salt and 0.10 N NaOH. At the end of the kinetic runs, the reaction mixtures were titrated against standard potassium hydrogen phthalate to secure the hydroxide concentration needed for calculating second-order rate constants.

Six identical kinetic runs with $9.80 \times 10^{-6}$ M pNPL (0.10 N NaOH, 25.0 °C) gave observed first-order rate constants of 2.37, 2.43, 2.58, 2.51, 2.44, and $2.44 \times 10^{-3}$ $s^{-1}$. To achieve such a high reproducibility it is necessary to add the substrate to the 0.10 N NaOH with a flattened stirring rod as described above. If the pNPL is introduced with a syringe followed by manual shaking of the cuvette, then the reproducibility of the rate constants is not nearly as good. We suspect that the latter procedure permits a sufficiently high local concentration of pNPL to cause precipitation of the solid. Four recrystallizations of *n*-butyltrimethylammonium iodide did not change the rate constant relative to that secured with twice-recrystallized material.

Rate constants for the hydrolysis of *p*-nitrophenyl acetate in the presence of additives were determined by using cuvettes filled with $4.5 \times 10^{-5}$ M ester, 0.05 M borate buffer (pH 10.00), and 0.0–0.5 M additive at 25.0 °C. The spectrophotometer was set at 400 nm and at the 0–2 absorbance scale.

## Results

Observed rate constants for basic hydrolysis of pNPL (0.10 N NaOH, 25.0 °C) decrease sharply and then level off, as the initial pNPL concentration increases from $10^{-6}$ to $10^{-5}$ M (see Figure 1 in ref 14). We attribute this to aggregation. All experiments in the present work were carried out at $9.80 \times 10^{-6}$ M pNPL where the second-order rate constant for saponification equals $2.45 \times 10^{-2}$ $M^{-1}$ $s^{-1}$. Ester is fully aggregated at $9.80 \times 10^{-6}$ M because the rate varies little with concentration above this level. First-order plots for systems with and without additives show a satisfactory linearity (Figure 2) as long as they are restricted to the first half-life. Linearity gives way to curvature beyond the first half-life owing to ultimate destruction of the aggregates as the ester hydrolyzes.

Hydrolysis rates increase upon adding quaternary ammonium salts ($RNMe_3^+X^-$) to the aqueous solutions of pNPL. Figure 3 illustrates the effect for 0.1–0.5 M *n*-propyltrimethylammonium iodide. One possible explanation for the rate enhancements, namely a medium effect at the reaction site, can be discarded owing to experiments with a nonaggregating ester, *p*-nitrophenyl acetate. The *p*-nitrophenyl acetate hydrolysis ($k_{obsd} = 2.32 \times 10^{-3}$ $s^{-1}$ at pH 10.05, 25.0 °C) is unperturbed by 0.45 M **2**, 0.25 M **9**, 0.24 M **16**, and 0.29 M **21** (see Table I for numbering of salts). We conclude that the salt-enhanced hydrolysis of pNPL, such as

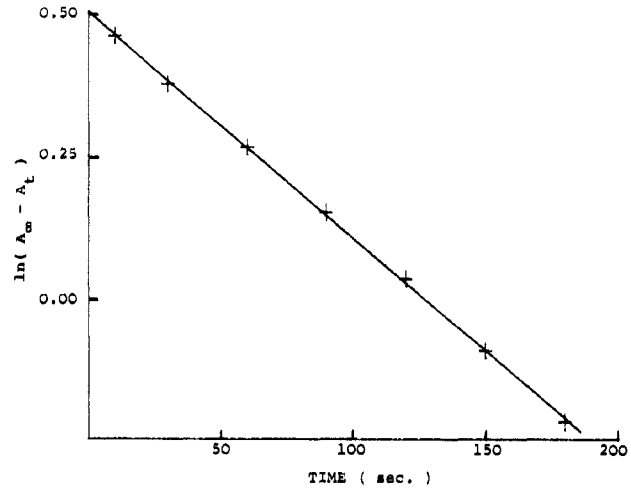



**Figure 2.** First-order plot for the hydrolysis of *p*-nitrophenyl laurate ($9.8 \times 10^{-6}$ M) at 25.0 °C in aqueous 0.10 N NaOH with 0.30 M *n*-propyltrimethylammonium iodide. Only the first half-life of the reaction is monitored.

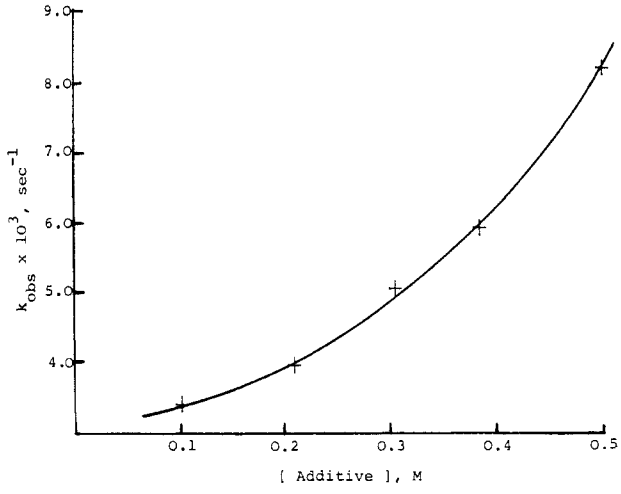



**Figure 3.** Observed rate constants for the hydrolysis of *p*-nitrophenyl laurate ($9.8 \times 10^{-6}$ M, 25.0 °C, 0.10 N NaOH) in the presence of varying concentrations of *n*-pentyltrimethylammonium iodide.

in Figure 3, stems from an interaction between the salts and the hydrocarbon chains of the laurate ester. Presumably, the salts disrupt or destroy the pNPL aggregates, thereby exposing ester groups to hydroxide.

Plots of $k_{obsd}$ vs. [salt], as in Figure 3, curve upward. In contrast, the log $k_{obsd}$ vs. [salt] plots are linear, a behavior observed for all 25 quaternary ammonium salts (Figure 4). Although linearity may be rationalized in terms of the Setschenow equation (a treatment of solubilization phenomena),[15] it is best to accept linearity here as a simple experimental observation.

If the salts do indeed interact with the laurate chains, then the slopes of log $k_{obsd}$ vs. $[RNMe_3^+X^-]$ should increase with the hydrophobicity of R. This was found to be the case. Thus, the slopes are 0.59, 0.73, 0.83, 0.97, 1.12, and 1.33 for R = methyl, ethyl, *n*-propyl, *n*-butyl, *n*-pentyl, and *n*-hexyl, respectively. (Note that we did not examine homologues higher than hexyl in order to minimize any possibility that the salt itself forms an aggregate.[16]) Encouraged by the obvious correlation between slope and hydrophobicity, we examined a whole family of R groups. Their slopes (henceforth called the microscopic hydrophobicity or "MH" value) are compiled in Table II. They range from 0.59 to 3.52 with an uncertainty of ±6%. In the next section, we use

(15) Long, F. A.; McDevit, W. F. Chem. Rev. **1952**, *51*, 119. See p 123.
(16) The cmc of hexyltrimethylammonium bromide is greater than 1 M. See: Osipow, L. I. *Surface Chemistry*; Reinhold: New York, 1962; p 165. The possibility of small submicellar aggregates cannot be excluded.

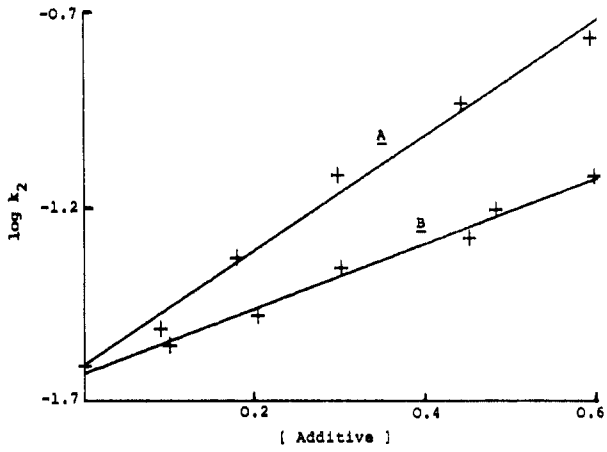



**Figure 4.** Least-squares plots of log $k_2$ for the hydrolysis of *p*-nitrophenyl laurate ($9.8 \times 10^{-6}$ M, 25.0 °C, 0.10 N NaOH) in the presence of varying amounts of additives (A = $C_6H_5NMe_3^+I^-$ and B = *n*-$C_3H_7NMe_3^+I^-$). The second-order rate constant $k_2 = k_{obsd}/[OH^-]$. MH values are obtained from the slopes of such plots.

**Table II.** Microscopic Hydrophobicity ("MH") Parameters for 25 Substituents

| no. | substituent | counterion | MH[a] |
|---|---|---|---|
| **1** | methyl | Br | 0.41 (0.59) |
| **2** | ethyl | I | 0.73 |
| **3** | *n*-propyl | I | 0.83 |
| **4** | isopropyl | I | 0.73 |
| **5** | *n*-butyl | I | 0.97 |
| **6** | *tert*-butyl | I | 0.88 |
| **7** | *n*-pentyl | I | 1.12 |
| **8** | 1-methylbutyl | I | 1.03 |
| **9** | *n*-hexyl | I | 1.33 |
| **10** | cyclopentyl | I | 0.96 |
| **11** | cyclohexyl | I | 1.13 |
| **12** | allyl | I | 0.67 |
| **13** | 2-propynyl | I | 0.76 |
| **14** | 3-butenyl | I | 0.78 |
| **15** | phenyl | I | 1.48 |
| **16** | benzyl | I | 1.60 |
| **17** | 4-chlorobutyl | I | 0.83 |
| **18** | 4-bromobutyl | I | 0.97 |
| **19** | 4-iodobutyl | I | 1.49 |
| **20** | 2-ethoxyethyl | Br | 0.73 (1.06) |
| **21** | 2-phenoxyethyl | Br | 1.85 (2.68) |
| **22** | *p*-methylbenzyl | Br | 1.86 (2.70) |
| **23** | *p*-fluorobenzyl | Br | 1.23 (1.78) |
| **24** | *p*-chlorobenzyl | Br | 2.14 (3.10) |
| **25** | *p*-bromobenzyl | Br | 2.43 (3.52) |

[a] Numbers in parentheses are MH values calculated for the iodide salts as described in the text.

the kinetically based scale to examine how branching, unsaturation, cyclization, aromaticity, halogenation, etc., affect hydrophobic association.

One cannot assume, of course, that the ionic head group of the salts has no effect on the aggregation properties of pNPL. For this reason, it was necessary to maintain a constant head group: $-NMe_3^+I^-$. Owing to solubility problems, however, compounds **1** and **20–25** in Table II could be examined only in the bromide form. MH values for these few cases were estimated with the aid of a correction factor equal to 1.45 (obtained from data on the bromide and iodide salts of **2** and **4**). Thus, an observed slope of 0.73 for (2-ethoxyethyl)trimethylammonium bromide (**20**) was multiplied by 1.45 to obtain an MH value of 1.06 for the corresponding iodide. Such indirectly calculated MH values are listed in parentheses in Table II.

## Discussion

Numerous investigators have confirmed our earlier observation that long-chain esters self-aggregate in water.[14] Guthrie[17] found

(17) Guthrie, J. P. *J. Chem. Soc., Chem. Commun.* **1972**, 897.

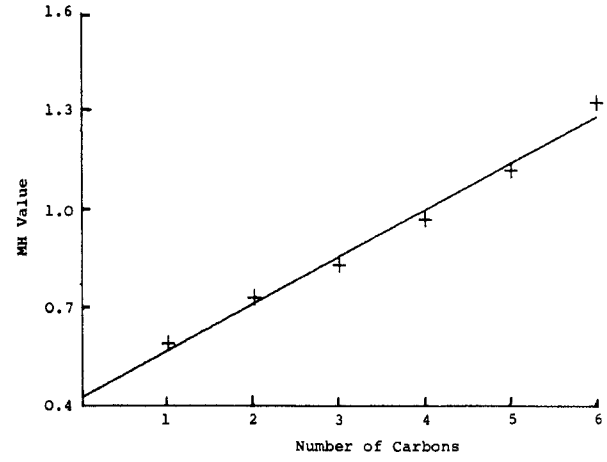



**Figure 5.** Plot of MH values (see Table II) vs. number of carbons in *n*-alkyl group (R) of $RNMe_3^+I^-$.

that *p*-nitrophenyl decanoate manifests a "critical concentration" of $1.6 \times 10^{-6}$ M; above this concentration the ester abruptly aggregates much like a conventional surfactant. Monomeric fatty esters (i.e., esters below their critical concentration) coil and thereby experience slightly reduced hydrolysis rates relative to short-chain analogues. Murakami and co-workers[18] used kinetic and tensiometric methods to determine critical concentrations for several long-chain esters. Hydrolyses taking place above critical concentrations presumably arise from low levels of monomer in equilibrium with the aggregates. Thus, inorganic salt can reduce the overall hydrolysis rate by driving monomer from the water into the aggregates (a type of "salting out"). Recently, Jiang and co-workers[19] provided compelling evidence that aggregation and coiling jointly retard the hydrolysis of long-chain esters.

Although there is no doubt that *p*-nitrophenyl laurate (pNPL) does indeed form aggregates in water, we know nothing about the structure of the aggregates. Attempts to determine aggregation numbers by light scattering failed because of the extremely low concentration at which one must operate ($<1 \times 10^{-5}$ M). Yet two points can be made with regard to aggregate structure: (1) pNPL aggregates certainly do not constitute a second phase ("microdroplets"); neither ourselves nor others have ever detected heterogeneity in aqueous pNPL solutions below $10^{-5}$ M. (2) While we would want to characterize the size and shape of pNPL aggregates were this feasible, the information is fortunately not needed. The key facts are that pNPL associates hydrophobically in water and that disruption of the resulting aggregates can be monitored kinetically.

As mentioned in the previous section, all 25 salts in Table II enhance pNPL hydrolysis. Several mechanisms for the salt effect seem possible: (1) The salts bind to the aggregates, causing them to swell or dissociate. This "deshields" the ester groups and exposes them to hydroxide ion. (2) The salts may drive the aggregate $\rightleftarrows$ monomer equilibrium to the right by binding to monomeric pNPL. Since monomer reacts much faster than aggregated material,[18] the rate increases. (3) The salts may enter the aggregates and effect a "phase-transfer" catalysis within the hydrocarbon microenvironment. Whatever the mechanism, it is clear that association between the R groups of the salts and the laurate of pNPL governs the kinetic perturbations. Thus, the MH values in Table II, secured from linear plots of log $k_{obsd}$ vs. $[RNMe_3^+X^-]$, reflect the lipophilicity of the R groups.

Let us now inspect various MH values in Table II:

(a) MH values increase uniformly as straight-chain Rs lengthen from methyl to *n*-hexyl (salts **1**, **2**, **3**, **5**, **7**, and **9**). A plot of MH vs. the number of carbons (Figure 5) is linear and can be used to estimate MH values for higher alkyl groups (e.g., MH of *n*-octyl = 1.57). Linearity is also achieved in a plot of log (activity) vs.

(18) Murakami, Y.; Aoyama, Y.; Kida, M. *J. Chem. Soc., Perkin Trans. 2* **1977**, 1947.

(19) Jiang, X.; Hui, Y.; Fan, W. *J. Am. Chem. Soc.* **1984**, *106*, 3839.

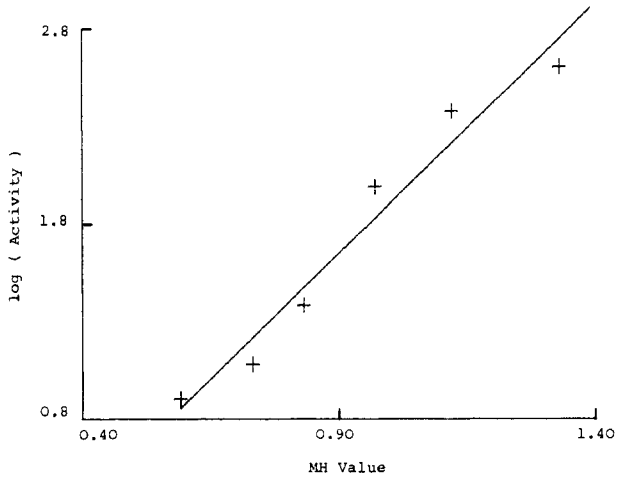
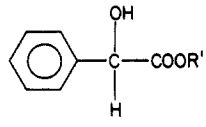



**Figure 6.** Least-squares plot of log (spasmolytic activity) for a homologous series of drugs bearing different R groups vs. MH values for the R groups (Table II). The structure of the drug is given in the text. Activity data taken from the following: Korolkovas, A. *Essentials of Molecular Pharmacology*; Wiley-Interscience: New York, 1970, p 77.

MH (Figure 6) for a spasmolytic drug, drawn below, where R′ is varied; drug potency clearly depends on hydrophobicity.




(b) Branching of R causes MH to diminish in three instances: *n*-propyl (0.83) and isopropyl (0.73); *n*-pentyl (1.12) and 1-methylbutyl (1.03); *n*-butyl (0.97) and *tert*-butyl (0.88). Although the branching effect seems real, it is small (only slightly greater than the experimental error in MH). Perhaps the smaller MH values for branched substituents are related to the hydration peculiarities of methyl groups discussed recently by Jorgensen et al.[20]

(c) Cyclization of a hydrocarbon chain reduces its hydrophobicity: compare *n*-pentyl (MH = 1.12) vs. cyclopentyl (MH = 0.96) and *n*-hexyl (MH = 1.33) vs. cyclohexyl (MH = 1.13).

(d) Unsaturation inserted into a saturated chain impairs hydrophobicity in all three cases examined: MH of *n*-propyl = 0.83 while MH of allyl = 0.67; MH of *n*-butyl = 0.97 while MH of 3-butenyl = 0.78; MH of *n*-propyl = 0.83 while MH of 2-propynyl = 0.76.

(e) Phenyl has a larger MH than cyclohexyl (1.48 vs. 1.13, respectively).

(f) Hydrophobic interaction for halides follows the order I > Br > Cl > F (see compounds **17–19** and **23–25**).

(g) An ether oxygen contributes to hydrophobicity but less so than a methylene. Thus, 2-ethoxyethyl (1.06) lies between *n*-butyl (0.97) and *n*-pentyl (1.12). Obviously, conclusions must be tentative since we have only one example and since the value of 1.06 for the 2-ethoxyethyl was obtained indirectly as described earlier.

(h) By far the largest MH value was obtained for *p*-bromobenzyl (MH = 3.52). Since the parent benzyl group has an MH of only 1.60, incorporating bromine has a huge effect (possibly due to dispersion forces). Even a *p*-chloro promotes hydrophobicity (increasing benzyl from 1.60 to 3.10; compare salts **16** and **24**). Halides would, therefore, seem to be logical candidates for enhancing transport of structurally nonspecific drugs (such as certain hypnotics, general anesthetics, insecticides, and disinfectants).

(i) 4-Bromobutyl and *n*-butyl are identical (MH = 0.97). Apparently, the "halide effect" applies only to aromatic systems.

(j) MH values and Hansch $\pi$ parameters correlate well ($r$ = 0.97) when substituents R of $RNMe_3^+X^-$ are saturated hydrocarbons (i.e., salts **1–7** and **11** for which $\pi$ parameters are available). An excellent correlation ($r$ = 0.99) exists between MHs of the *p*-halobenzyl salts (**23–25**) and the $\pi$ parameters for fluoro, chloro, and bromo (0.14, 0.71, and 0.86, respectively).[2] MH and $\pi$ for the aromatics do not correspond. For example, phenyl and *tert*-butyl have nearly identical $\pi$ values, whereas the phenyl MH is nearly 70% greater than that of *tert*-butyl. The origin of this high aromatic lipophilicity, as measured here kinetically, is not clear. An interaction between the R groups and the pNPL head groups seems unlikely (unless aggregation specifically promotes such an effect) because pNPA hydrolysis is hardly perturbed by aromatic quaternary ammonium salts. There is, of course, no need to expect a general MH/$\pi$ correlation since the two parameters are based on entirely different measurements.

(20) Jorgensen, W. L.; Gao, J.; Ravimohan, C. *J. Phys. Chem.* **1985**, *89*, 3470.

## Summary

We have developed a method to assess hydrophobic character which exploits the fact that $RNMe_3^+X^-$ salts disrupt or destroy *p*-nitrophenyl laurate aggregates. Microscopic hydrophobicity or "MH" parameters were evaluated for 25 substituents R. The disadvantages of the method include the following: (1) Most of the necessary salts are commercially unavailable (and often unknown). The synthetic work that must obviously precede the MH determinations impedes, at least with us, a massive screening of R groups. (2) Hydrophilic substituents (e.g., $-CH_2COOH$) that do not perturb the aggregates will not provide MH values. (3) Nucleophilic substituents (e.g., those possessing an amino group) can react covalently with the long-chain ester; this interferes with MH studies. (4) Comparisons among the substituents are made by using solutions with an ionic strength of 0.2–0.7 as opposed to pure water. Admittedly, these are serious drawbacks. Yet the approach can, as we have just seen, be used to decipher the role of branching, unsaturation, aromaticity, halogenation, etc., in hydrophobic behavior. And since MH parameters are based on the binding of one molecule to another (rather than partitioning between solvents), they may better model biological processes where intermolecular association is taking place.

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