ELSEVIER

Journal of Chromatography B, 745 (2000) 117–126

JOURNAL OF CHROMATOGRAPHY B

www.elsevier.com/locate/chromb

# Rapid method for estimating the octanol–water partition coefficient (log $P_{ow}$) by microemulsion electrokinetic chromatography

Salwa K. Poole*, Douglas Durham, Christopher Kibbey

*Chemistry Department, Parke-Davis Pharmaceutical Research, Division of Warner-Lambert Company, 2800 Plymouth Road, Ann Arbor, MI 48105, USA*

**Abstract**

Several surfactant systems were evaluated based on their system constants determined by the solvation parameter model for the design of a surrogate chromatographic model for the rapid estimation of octanol–water partition coefficient (log $P_{ow}$) by microemulsion electrokinetic chromatography. The system constant ratios responsible for the log $P_{ow}$ partition system are (nearly) the same as those for the microemulsion system containing sodium dodecyl sulfate (1.4% w/v), butan-1-ol (8% v/v) and heptane (1.2% v/v). Neutral and basic compounds are analyzed using a fused-silica capillary column with a 50 m*M* sodium phosphate–sodium borate (3:2) buffer at pH 10. Weakly acid compounds require the use of sulfonated silica capillary column and a 50 m*M* sodium phosphate buffer at pH 3. For 29 varied neutral and weakly basic compounds the average error between log $P_{ow}$ estimated using MEEKC and literature values was $\pm 0.12$ over a log $P_{ow}$ range from 0.3 to 5.8. 

*Keywords:* Octanol–water partition coefficient; Microemulsion electrokinetic chromatography

## 1. Introduction

The octanol–water partition coefficient (log $P_{ow}$) is widely used as a general measure of lipophilicity, and as a parameter to predict transport properties across a cell membrane in drug discovery research [1,2]. A number of direct methods including shake-flask, stir-flask, two-phase titration, generator column, counter-current chromatography and flow injection extraction have been described for the determination of log $P_{ow}$ [3,4]. In general, these methods are time consuming, labor intensive, require significant amounts of pure compounds, have a limited dynamic range, and are difficult to automate. Faster and more economical methods are desirable to support high throughput screening of candidate drug substances in the pharmaceutical industry. Indirect chromatographic methods for estimating log $P_{ow}$ are faster and easier to automate than traditional shake-flask and titration methods. In addition, chromatographic methods do not require pure materials and provide reliable data for small sample sizes. There are now numerous reports on the application of thin-layer chromatography [5–7], reversed-phase column liquid chromatography [8–14], and micellar electrokinetic chromatography [14–20] for the estimation of log $P_{ow}$. These methods are based on the construction of a correlation model between a retention property characteristic of the solute and the chromatographic system for a training set of solutes with known experimental octanol–water partition coefficients. Then further measurements of the chro-

*Corresponding author. Tel.: +1-734-622-3077; fax: +1-734-622-2716.

*E-mail address:* pooles@aa.wl.com (S.K. Poole).

0378-4347/00/$ – see front matter 
PII: S0378-4347(00)00072-4

matographic retention property in the same system are used to estimate log $P_{ow}$ values for other compounds. A common feature of these models is that they produce acceptable estimates for compounds with similar functional groups, or belonging to a homologous series. Results are generally poor when a varied group of compounds are analyzed without first sorting the compounds by class or functional group. These aspects of the more common chromatographic methods limit their scope for the rapid estimation of log $P_{ow.}$ Error in the estimated log $P_{ow}$ arise because common solvated sorbents for reversed-phase liquid chromatography are poor surrogate models for the wet octanol [13,14,21–24]. In general, reversed-phase chromatographic sorbents lack sufficient hydrogen-bond basicity and are more dipolar/polarizable than desired to be good models for wet octanol.

Systematic studies to define a suitable surrogate chromatographic model for log $P_{ow}$ are based on defining the contribution of solute size and intermolecular interactions to the transfer of solutes from water to wet octanol and identifying a chromatographic system with (nearly) identical distribution properties [14,25,26]. The solvation parameter model, as set out below, is the most useful approach for this purpose

$$\log \mathrm{SP} = c + mV_X + rR_2 + s\pi_2^H + a\Sigma\alpha_2^H + b\Sigma\beta_2^0 \quad (1)$$

The model is made up of product terms representing solute properties (descriptors) and system properties. Each product term represents the contribution of defined intermolecular interactions to the correlated solute property, log $P_{ow}$ or the retention factor, log $k$, in chromatography. The solute descriptors are McGowan's characteristic volume $V_X$, excess molar refraction $R_2$, the solute's dipolarity/dipolarizability $\pi_2^H$, and the solute's effective hydrogen-bond acidity and hydrogen-bond basicity, $\Sigma\alpha_2^H$ and $\Sigma\beta_2^0$, respectively. Solute descriptors are available for about 4000 compounds and additional values can be obtained by parameter estimates or experiment [26,27].

The system constants in Eq. (1) are unambiguously defined. The $r$ constant refers to the difference in capacity of the two phases to interact with solute n- or π-electrons. The $s$ constant to the difference in capacity of the two phases to take part in dipole-type interactions. The $a$ constant is a measure of the difference in hydrogen-bond basicity of the two phases. The $b$ constant is a measure of the difference in hydrogen-bond acidity of the two phases. The $m$ constant is a measure of the relative ease of forming a cavity for the solute in the two phases together with the difference in solute–solvent dispersion interactions in the two phases. The system constants are obtained by multiple linear regression analysis of experimental log SP values for a varied group of solutes with known descriptors [28].

Abraham obtained the following solvation parameter model for log $P_{ow}$ [26]

$$\log P_{ow} = 0.088 + 3.841V_X + 0.562R_2 - 1.054\pi_2^H + 0.034\Sigma\alpha_2^H - 3.460\Sigma\beta_2^0 \quad (2)$$

$n = 613$, $\rho = 0.997$, SE = 0.116, $F = 23162$

where $n$ is the number of solutes, $\rho$ is the overall correlation coefficient, SE is the standard error in the estimate and $F$ is Fischer's statistic. The system constants of this model are significantly different to those observed for reversed-phase liquid chromatographic systems but more general agreement is indicated for micellar phases used in micellar electrokinetic chromatography [14,27–31]. Abraham et al. [32] indicated that the microemulsion containing sodium dodecyl sulfate (1.44% w/w), n-butanol (6.49% w/w) and n-heptane (0.82% w/w) should provide a reasonable surrogate chromatographic model for log $P_{ow}$ based on a comparison of system constant ratios. This system was used by Gluck et al. [33] for the estimation of log $P_{ow}$ values for 23 neutral and acidic compounds (pH = 1.19) and 13 neutral and basic compounds (pH = 12). The average error in the estimate of log $P_{ow}$ was ±0.4 for log $P_{ow}$ values from −1.0 to 4.4. At low pH the electroosmotic flow in fused-silica capillary columns is very slow and the micellar migration time increases to an unacceptable value. Lin and Pietrzyk [34] demonstrated that the electroosmotic flow of fused-silica capillary columns coated with a sulfonic acid polymer is independent of pH and more suitable for separations at low pH than native fused-silica capillary columns.

In this paper micellar and microemulsion pseudo-

phases are evaluated for the estimation of log $P_{ow}$ using acidic and basic buffers in an attempts to identify set of conditions amenable to the rapid estimation of log $P_{ow}$. The proposed method is validated for a large set of variegated compounds with diversity of structure.

## 2. Experimental

Sodium cholate, sodium dodecyl sulfate, and all compounds used as solutes were obtained from Aldrich Chemical Co. (Milwaukee, WI, USA). Sodium borate, sodium phosphate, 1-butanol and methanol were obtained from Mallinkrodt (Phillipsburg, NJ, USA).

The sodium cholate containing buffer was prepared by adding sufficient sodium cholate by weight to 50 m*M* sodium phosphate buffer, adjusted to pH = 7, 8, and 9 by adding concentrated sodium hydroxide (1 *M*), to give a final concentration of 80 m*M*. The sodium dodecyl sulfate microemulsion was prepared by adding sodium dodecyl sulfate (1.44 g) and 1-butanol (8 ml) to 90 ml of the appropriate buffer with ultrasonic mixing. Heptane (1.2 ml) was added with mixing to the clear solution. The final solution was made up to 100 ml with buffer and allowed to stand at room temperature until the solution became clear (about 1 h). The pH 10 buffer for preparation of the microemulsion was prepared by mixing 50 ml of 50 m*M* sodium borate with 75 ml of 50 m*M* sodium phosphate. For the pH 3 buffer phosphoric acid (85%) was added dropwise to 50 m*M* sodium phosphate.

All retention factor measurements were made on a Beckman P/ACE System MDQ (Fullerton, CA, USA) equipped with a photodiode array detector operated at 214 nm. The applied voltage was adjusted to 20–30 kV to maintain the current below 100 μA. The inlet vial was pressurized to 0.2 p.s.i. to reduce retention times. All retention measurements were made at 30°C.

Uncoated fused-silica capillary columns, 72 cm long (effective length 62 cm) and 50 μm I.D. were purchased from Perkin-Elmer (Norwalk, CT, USA) and were used for all measurements except at pH 3. For the latter sulfonic acid coated fused-silica capillary columns 50 cm long (effective length 40 cm) and 50 μm I.D. were purchased from Scientific Resources (Easontown, NJ, USA). Windows for on-column detection were prepared using a frit burner from Innovatech (Stevenage, UK). Prior to use the uncoated fused-silica capillary columns were conditioned by flushing with sodium hydroxide (1.0 *M*) for 15 min, followed by water for 15 min and finally with the buffer for 15 min. The sulfonic acid coated fused-silica capillaries were conditioned by flushing with the running buffer for 15 min. Prior to each separation the capillaries were back flushed at 75 p.s.i. with the running buffer for 2 min. The vial caps should be kept dry at all times to avoid arcing, which can result in column breakage.

Standard solutions were made up in methanol (1–2 mg/ml). 25 μl of standard solution was mixed with 50 μl of dodecanophenone in methanol (15 mg/ml) and diluted with 335 μl of the micelle or microemulsion solution made up in water (prepared as described above excluding the buffer components). Samples were introduced into the capillary by applying a pressure of 1 p.s.i. for 6 s. The retention factor, *k*, was calculated using Eq. (3)

$$k = (t_R - t_{eo})/(1 - t_R/t_{mc})t_{eo} \quad (3)$$

where $t_{eo}$ is the migration time of the electroosmotic flow marker (methanol), $t_{mc}$ is the migration time of the micelle marker (dodecanophenone), and $t_R$ is the solute migration time.

The solute descriptors for the solvation parameter model [25,28,35–38] and log $P_{ow}$ [38,39] were taken from several sources and are tabulated in Table 1 for the reader's convenience. Multiple linear regression analysis and statistical tests were performed on a Gateway G6 −333 personal computer (Sioux City, ND, USA) using the program SPSS/PC V. 9 (SPSS, Chicago, IL, USA).

## 3. Results and discussion

From the several hundred chromatographic systems that have been characterized by the solvation parameter model only six have properties similar to the octanol–water distribution system, and five of these are micellar or microemulsion systems [14,23]. The two systems most likely to provide an adequate surrogate chromatographic model are the sodium

Table 1
Solute descriptors used in the solvation parameter model and log $P_{OW}$ for the evaluation solute set

| Compound | Solute descriptor | | | | | log $P_{OW}$ |
|---|---|---|---|---|---|---|
| | $V_x$ | $R_2$ | $\pi$ | $\Sigma\alpha$ | $\Sigma\beta$ | |
| Acridine | 1.413 | 2.356 | 1.32 | 0.0 | 0.58 | 3.40 |
| Acetophenone | 1.014 | 0.818 | 1.01 | 0.0 | 0.48 | 1.58 |
| Acetylsalicylic acid | 1.288 | 0.781 | 0.80 | 0.49 | 1.00 | 1.19 |
| Aldosterone | 2.689 | 2.021 | 3.47 | 0.40 | 1.90 | 1.08 |
| Anisole | 0.916 | 0.708 | 0.75 | 0.0 | 0.29 | 2.11 |
| Aniline | 0.816 | 0.955 | 0.96 | 0.26 | 0.50 | 0.84 |
| Anthracene | 1.454 | 2.290 | 1.34 | 0.0 | 0.26 | 4.45 |
| Antipyrene | 1.550 | 1.320 | 1.50 | 0.0 | 1.48 | 0.38 |
| Acetaminophen | 1.172 | 1.060 | 1.78 | 1.09 | 0.81 | 0.51 |
| Benzamide | 0.973 | 0.990 | 1.5 | 0.49 | 0.67 | 0.64 |
| Benzyl alcohol | 0.916 | 0.803 | 0.87 | 0.39 | 0.56 | 1.01 |
| Benzaldehyde | 0.873 | 0.820 | 1.00 | 0.0 | 0.39 | 1.48 |
| Bromobenzene | 0.891 | 0.882 | 0.73 | 0.0 | 0.09 | 2.99 |
| Butylbenzene | 1.280 | 0.600 | 0.51 | 0.0 | 0.15 | 4.38 |
| Caffeine | 1.363 | 1.400 | 1.55 | 0.0 | 1.34 | −0.16 |
| Cortisone | 2.755 | 1.960 | 3.50 | 0.36 | 1.84 | 1.42 |
| 4-Chloroaniline | 0.939 | 1.060 | 1.13 | 0.30 | 0.35 | 1.88 |
| 4-Chlorotoluene | 0.979 | 0.705 | 0.67 | 0.0 | 0.05 | 3.33 |
| 2,4-Dinitrophenol | 1.124 | 1.20 | 1.50 | 0.10 | 0.55 | 2.36 |
| Diphenylamine | 1.424 | 0.700 | 0.88 | 0.60 | 0.38 | 3.50 |
| Coumarin | 1.062 | 1.06 | 1.79 | 0.0 | 0.46 | 1.60 |
| 1,3-Dichlorobenzene | 0.961 | 0.847 | 0.73 | 0.0 | 0.02 | 3.53 |
| Ethylbenzoate | 1.214 | 0.689 | 0.85 | 0.0 | 0.46 | 2.64 |
| Estradiol | 2.199 | 1.800 | 3.30 | 0.88 | 0.95 | 2.69 |
| Estrone | 2.156 | 1.73 | 3.10 | 0.56 | 0.91 | 2.76 |
| Eugenol | 1.354 | 0.946 | 0.99 | 0.22 | 0.51 | 2.99 |
| Fluoranthene | 1.585 | 2.377 | 1.53 | 0.0 | 0.20 | 4.50 |
| Hydrocortisone | 2.798 | 2.030 | 3.49 | 0.71 | 1.87 | 1.55 |
| Ibuprofen | 1.777 | 0.700 | 0.92 | 0.60 | 0.60 | 3.50 |
| Imipramine | 2.402 | 1.480 | 1.75 | 0.0 | 1.19 | 3.49 |
| Indazole | 0.905 | 1.180 | 1.35 | 0.54 | 0.30 | 1.77 |
| Iodobenzene | 0.974 | 1.188 | 0.82 | 0.0 | 0.12 | 3.25 |
| Indole | 0.946 | 1.200 | 1.12 | 0.44 | 0.31 | 2.14 |
| 4-Methylbenzamide | 1.114 | 0.990 | 1.50 | 0.49 | 0.65 | 1.18 |
| Methylbenzoate | 1.073 | 0.733 | 0.85 | 0.0 | 0.48 | 2.12 |
| 1-Methylnaphthalene | 1.226 | 1.344 | 0.90 | 0.0 | 0.20 | 3.87 |
| 4-Methylphenol | 0.916 | 0.820 | 0.87 | 0.57 | 0.31 | 1.94 |
| Naphthalene | 1.085 | 1.340 | 0.92 | 0.0 | 0.20 | 3.37 |
| Nicotine | 1.371 | 0.865 | 0.75 | 0.0 | 1.14 | 1.17 |
| 4-Nitrobenzamide | 1.147 | 1.250 | 2.17 | 0.75 | 0.60 | 1.93 |
| 4-Nitroaniline | 0.991 | 1.220 | 1.91 | 0.42 | 0.38 | 1.39 |
| 1-Nitronaphthalene | 1.260 | 1.600 | 1.51 | 0.0 | 0.29 | 3.19 |
| 4-Nitrophenol | 0.949 | 1.070 | 1.72 | 0.82 | 0.26 | 1.91 |
| Phenanthrene | 1.454 | 2.055 | 1.29 | 0.0 | 0.26 | 4.46 |
| Phenol | 0.7751 | 0.805 | 0.89 | 0.6 | 0.31 | 1.46 |
| Phenylacetate | 1.072 | 0.661 | 1.13 | 0.0 | 0.54 | 1.41 |
| Prednisolone | 2.755 | 2.210 | 3.10 | 0.71 | 1.92 | 1.62 |
| Pregnenolone | 2.665 | 1.360 | 3.29 | 0.32 | 1.18 | 3.13 |
| Progesterone | 2.622 | 1.450 | 3.29 | 0.0 | 1.14 | 3.26 |
| Pyrazine | 0.634 | 0.629 | 0.95 | 0.0 | 0.61 | −0.26 |
| Pyrene | 1.585 | 2.808 | 1.71 | 0.0 | 0.29 | 4.88 |
| Pyrrole | 0.577 | 0.613 | 0.73 | 0.41 | 0.29 | 0.75 |
| Quinoline | 1.044 | 1.268 | 0.97 | 0.0 | 0.54 | 2.03 |
| Resorcinol | 0.834 | 0.980 | 1.00 | 1.10 | 0.58 | 0.80 |
| Salicylic acid | 0.990 | 0.890 | 0.70 | 0.72 | 0.41 | 2.26 |
| Toluene | 0.857 | 0.601 | 0.52 | 0.0 | 0.14 | 2.69 |
| Valerophenone | 1.437 | 0.800 | 0.95 | 0.0 | 0.50 | 3.11 |

Table 2
Retention factors obtained by micellar and microemulsion electrokinetic chromatography

| Compound | Logarithm of the retention factor (log $k$) | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | SC7 | SC8 | SC9 | Em7 | Em8 | Em10 | EM12 | EM3 |
| Acridine | | 0.777 | | | 1.062 | 0.972 | 1.069 | |
| Acetophenone | −0.328 | −0.323 | −0.0335 | 0.067 | 0.123 | 0.057 | 0.07 | 0.046 |
| Acetylsalicylic Acid | | −0.426 | | | −0.255 | | | −0.255 |
| Aldosterone | | −0.0103 | | | 0.231 | 0.217 | 0.261 | 0.238 |
| Anisloe | −0.060 | −0.040 | −0.038 | 0.348 | 0.465 | 0.327 | 0.361 | 0.360 |
| Aniline | −0.789 | −0.718 | −0.732 | −0.331 | −0.432 | −0.341 | −0.315 | |
| Anthracene | 1.333 | 1.693 | 1.654 | 2.028 | 2.157 | 1.992 | 2.145 | 1.980 |
| Antipyrene | −0.714 | −0.687 | −0.686 | −0.416 | −0.439 | −0.439 | −0.418 | −0.344 |
| Acetaminophen | −0.503 | −0.529 | −0.300 | | −0.631 | | | −0.656 |
| Benzamide | −0.649 | −0.611 | −0.626 | −0.363 | −0.370 | −0.384 | −0.367 | −0.379 |
| Benzyl Alcohol | −0.667 | −0.642 | −0.641 | −0.235 | −0.166 | −0.234 | −0.239 | −0.269 |
| Benzaldehyde | −0.492 | −0.466 | −0.459 | −0.051 | 0.0215 | −0.077 | −0.049 | 0.060 |
| Bromobenzene | 0.713 | 0.651 | 0.745 | 0.826 | 1.098 | 0.948 | 1.296 | 0.907 |
| Butylbenzene | 1.326 | | 1.626 | 1.960 | 1.691 | 1.460 | 1.987 | |
| Caffeine | −0.549 | | | −0.637 | −0.732 | −0.700 | −0.655 | |
| Cortisone | 0.073 | 0.0484 | 0.129 | | 0.273 | 0.274 | 0.283 | 0.268 |
| 4-Chloroaniline | 0.018 | −0.014 | 0.0497 | 0.385 | 0.372 | 0.333 | 0.400 | |
| 4-Chlorotoluene | 0.919 | 0.892 | 0.980 | 0.923 | 1.365 | 1.228 | 0.843 | |
| Coumarin | | | | | | | | −0.094 |
| 2,4-Dinitrophenol | | | | | | | | 0.337 |
| Dipheylamine | 0.913 | 0.801 | 0.891 | 1.234 | 1.243 | 1.135 | 1.291 | |
| 1,3-Dichlorobenzene | 1.098 | 0.965 | 1.059 | 1.340 | 1.518 | 1.328 | 1.388 | 1.279 |
| Ethylbenzoate | 0.359 | 0.346 | 0.400 | 0.744 | 0.859 | 0.742 | 0.780 | 0.704 |
| Estradiol | | 1.074 | 1.101 | | 1.128 | | | 1.116 |
| Estrone | | 1.243 | 1.273 | | 1.227 | 1.110 | 0.566 | 1.020 |
| Eugenol | | 0.704 | | | 0.761 | | | 1.050 |
| Fluoranthene | 1.751 | 1.708 | 1.672 | 2.213 | 2.226 | 2.241 | | 2.264 |
| Hydrocortisone | 0.176 | 0.142 | 0.192 | | 0.360 | 0.362 | 0.376 | 0.381 |
| Ibuprefen | | | | | | | | 1.499 |
| Imipramine | | | | | 1.501 | 1.447 | 1.966 | |
| Indazole | | −0.101 | | | 0.230 | 0.194 | 0.267 | |
| Iodobenzene | 0.949 | 0.886 | 1.004 | 1.167 | 1.281 | 1.139 | 1.443 | 1.133 |
| Indole | | 0.042 | | | 0.400 | 0.329 | 0.395 | |
| 4-Methylbenzamide | −0.521 | −0.522 | −0.504 | −0.284 | −0.317 | −0.316 | −0.267 | −0.316 |
| Methylbenzoate | 0.037 | 0.0136 | 0.0635 | 0.414 | 0.497 | 0.403 | 0.430 | 0.390 |
| 1-Methylnaphthalene | | 1.334 | | 1.726 | 1.827 | 1.539 | 1.683 | 1.580 |
| 4-Methylphenol | −0.108 | −0.105 | −0.072 | 0.212 | 0.277 | 0.297 | 0.610 | 0.185 |
| Naphthalene | 1.088 | 0.931 | 1.047 | | 1.343 | 1.195 | 1.561 | 1.144 |
| Nicotine | | | | | −0.487 | −0.157 | −0.048 | |
| 4-Nitroaniline | | −0.200 | | | −0.120 | 0.025 | 0.272 | |
| 4-Nitrobenzamide | | | | | | | | 0.173 |
| 1-Nitronaphthalene | | 0.896 | | | 1.156 | 1.1307 | 1.114 | 1.034 |
| 4-Nitrophenol | | | | | | | | 0.130 |
| Phenanthrene | 1.345 | 1.657 | 1.660 | 1.941 | 2.060 | 1.930 | 2.000 | 1.871 |
| Phenol | −0.491 | −0.442 | −0.271 | −0.168 | −0.08 | | | −0.206 |
| Phenylacetate | −0.463 | −0.451 | −0.235 | −0.207 | 0.078 | 0.078 | 0.0840 | 0.006 |
| Prednisolone | 0.2028 | | | | 0.384 | 0.379 | 0. 392 | 0.368 |
| Pregnenolone | | 1.424 | 1.515 | | 1.712 | 1.274 | 1.695 | 1.563 |
| Progesterone | | 1.134 | | | 1.599 | 1.474 | 1.443 | 1.312 |
| Pyrazine | −1.167 | −1.109 | −1.156 | −0.820 | −0.946 | −0.928 | −0.821 | |
| Pyrene | 1.763 | 1.783 | 1.912 | 2.174 | 2.249 | 2.1176 | | 2.226 |
| Pyrrole | −0.989 | −0.929 | −0.953 | −0.551 | −0.554 | −0.553 | −0.498 | |
| Quinoline | | | | | 0.324 | 0.279 | 0.340 | |
| Resorcinol | −0.459 | −0.416 | −0.295 | −0.487 | −0.462 | | | −0.536 |
| Salicylic acid | | | | | | | | 0.409 |
| Toluene | 0.446 | 0.359 | 0.382 | 0.848 | 1.042 | 0.713 | 0.958 | 0.681 |
| Valerophenone | 0.773 | | | 1.065 | 1.229 | 1.056 | 1.122 | 1.009 |

SC, Sodium cholate at pH 7, 8 and 9; EM, emulsion at pH 3, 7, 8, 10, and 12.

dodecyl sulfate/butanol/heptane microemulsion and the sodium cholate micelle systems. The influence of pH on these systems was studied because it might influence the solvation properties of the pseudo-stationary phase, it will influence the available migration window, and will also affect the applicability of the system for the estimation of log $P_{ow}$ for weak acids and bases. In an ionized form solutes are subject to additional electrophoretic migration and electrostatic interactions with the charged components of the separation system not considered by the solvation parameter model and are not expected to yield meaningful estimates of log $P_{ow.}$

To evaluate the retention properties of the surrogate chromatographic systems a group of compounds of sufficient number and variety to provide a good statistical model and at the same time representative of the general model for log $P_{ow}$, Eq. (2), were selected (see Table 1). To establish that these compounds are representative of the larger data set used to characterize the octanol–water partition system the following model was obtained

$$\log P_{ow} = \underset{(0.06)}{0.29} + \underset{(0.09)}{3.51}V_X + \underset{(0.05)}{0.55}R_2 - \underset{(0.06)}{1.00}\pi_2^H - \underset{(0.08)}{3.31}\Sigma\beta_2^0 \quad (4)$$

$n = 51$, $\rho = 0.994$, SE = 0.15, $F = 721$.

There is good agreement between Eqs. (2) and (4) and the model obtained is statistically sound indicating that the smaller data set provides an adequate representation of the information in the larger data set. Where the number of solutes in tables indicates smaller subsets they have been tested in the same way and shown to be in good agreement with Eq. (2).

The retention properties for the evaluation solute set were determined for the microemulsion and micelle system at different pH values and are summarized in Table 2. These results were fit to the solvation parameter model to provide the system constants summarized in Table 3. All models are statistically sound and the system constants make chemical sense. In order to provide an adequate correlation model for log $P_{ow}$ and the retention factor it is not necessary that the chromatographic system and the octanol–water distribution system have identical system constants. It is sufficient that the ratios of the system constants are (nearly) identical when normalized by division with the $m$ (or another) system constant [14,21–26]. The system constant

Table 3
System constants for different micellar phases and pH[a]

| System | pH | System constants | | | | | Statistics[b] | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | $m$ | $r$ | $s$ | $b$ | $c$ | $\rho$ | SE | $F$ | $n$ |
| SC | 7 | 2.135 | 0.537 | −0.934 | −1.419 | −1.120 | 0.965 | 0.23 | 103 | 35 |
| | | (0.174) | (0.095) | (0.161) | (0.164) | (0.108) | | | | |
| | 8 | 2.173 | 0.324 | −0.392 | −2.052 | −1.229 | 0.978 | 0.17 | 220 | 45 |
| | | (0.125) | (0.064) | (0.08) | (0.103) | (0.074) | | | | |
| | 9 | 2.251 | 0.254 | −0.358 | −2.143 | −1.173 | 0.974 | 0.21 | 139 | 35 |
| | | (0.165) | (0.089) | (0.125) | (0.151) | (0.100) | | | | |
| Emulsion | 7 | 2.391 | 0.529 | −0.972 | −1.699 | −0.909 | 0.989 | 0.15 | 286 | 31 |
| | | (0.139) | (0.111) | (0.203) | (0.144) | (0.141) | | | | |
| | 8 | 2.387 | 0.311 | −0.516 | −2.259 | −0.869 | 0.985 | 0.16 | 363 | 51 |
| | | (0.090) | (0.055) | (0.059) | (0.078) | (0.063) | | | | |
| | 10 | 2.239 | 0.369 | −0.511 | −1.968 | −0.845 | 0.990 | 0.12 | 471 | 45 |
| | | (0.073) | (0.042) | (0.050) | (0.0673) | (0.051) | | | | |
| | 12 | 2.371 | 0.315 | −0.666 | −2.015 | −0.735 | 0.962 | 0.23 | 119 | 43 |
| | | (0.137) | (0.095) | (0.095) | (0.121) | (0.102) | | | | |
| | 3 | 2.164 | 0.405 | −0.501 | −2.007 | −0.905 | 0.984 | 0.14 | 283 | 42 |
| | | (0.097) | (0.058) | (0.057) | (0.087) | (0.066) | | | | |

[a] SC, sodium cholate; Emulsion, 1.44% (w/v) sodium dodecyl sulfate, 8% (v/v) butanol and 1.2% (v/v) heptane

[b] $\rho$, correlation coefficient; SE, standard error in the estimate; $F$, Fischer F-statistic; $n$, number of solutes; and number in parentheses indicate the standard deviation in the coefficient.

ratios together with the difference in the system constant ratios for the chromatographic and octanol–water distribution systems (Δ) are summarized in Table 4. The system constant ratios are similar for sodium cholate at pH = 8 and 9, for the sodium dodecyl sulfate microemulsion at pH = 3, 8, and 10, and for the octanol–water distribution system. This is as required if a surrogate chromatographic model for the octanol–water distribution system for neutral and weakly acidic and basic compounds is to be developed. Sodium cholate is a weak acid and at low pH is unable to function as a charged micellar phase. The strong organic acid, sodium dodecyl sulfate, however, is completely ionized at pH 3 and thus behaves in the required manner. For both sodium cholate and the sodium dodecyl sulfate microemulsion at pH = 7 there is a significant change in the hydrogen-bond acidity and capacity of the pseudo-stationary phases for dipole-type interactions. These changes render this pH unsuitable for estimating log $P_{ow}$. At pH 12 there is a small change in the system properties for the sodium dodecyl sulfate microemulsion compared to pH 8 and 10. The correlation in system properties between octanol–water and the microemulsion system is better at pH 10 than at pH 12. In addition, the migration window at pH 12 is smaller than that at pH 8 and 10, which adversely affect the resolution of hydrophobic compounds.

In summary, sodium cholate is suitable for estimating log $P_{ow}$ for neutral and weakly basic compounds but unsuitable for weak acid compounds. The sodium dodecyl sulfate microemulsion is suitable for estimating log $P_{ow}$ values for neutral and weakly acidic and basic compounds at pH 3 and pH 10, respectively. The sulfonic acid coated capillary provides adequate electroosmotic flow at pH 3 without influencing the distribution properties of the microemulsion for neutral and weakly acidic compounds. Given the similarity in distribution properties between sodium cholate micelles and the sodium dodecyl sulfate microemulsion and the more favorable operating conditions for the microemulsion for acid conditions, this microemulsion system was selected for further evaluation at pH 3 and pH 10

First of all the correlation plot of log $P_{ow}$ against log $k$ for 45 neutral and basic compounds run in the

Table 4
System constant ratios for chromatographic models of the octanol–water distribution system. (Δ = difference in system constant ratios for octanol–water distribution and chromatographic system)

| System | pH | System constant ratios | | |
|---|---|---|---|---|
| | | $r/m$ | $s/m$ | $b/m$ |
| Octanol–Water | | 0.157 | −0.284 | −0.943 |
| SC | 7 | 0.252 | −0.437 | −0.665 |
| Δ | | 0.095 | 0.153 | 0.278 |
| SC | 8 | 0.149 | −0.180 | −0.944 |
| Δ | | 0.008 | 0.104 | 0.001 |
| SC | 9 | 0.113 | −0.159 | −0.952 |
| Δ | | 0.044 | 0.125 | 0.009 |
| Emulsion | 7 | 0.221 | −0.406 | −0.711 |
| Δ | | 0.064 | 0.123 | 0.232 |
| Emulsion | 8 | 0.130 | −0.216 | −0.946 |
| Δ | | 0.027 | 0.07 | 0.003 |
| Emulsion | 10 | 0.173 | −0.239 | −0.920 |
| Δ | | 0.016 | 0.045 | 0.023 |
| Emulsion | 12 | 0.133 | −0.281 | −0.849 |
| Δ | | 0.002 | 0.003 | 0.093 |
| Emulsion | 3 | 0.187 | −0.232 | −0.927 |
| Δ | | 0.030 | 0.053 | 0.016 |

SC, Sodium cholate; Emulsion, 1.44% (w/v) sodium dodecyl sulfate, 8% (v/v) butanol and 1.2% (v/v) heptane.

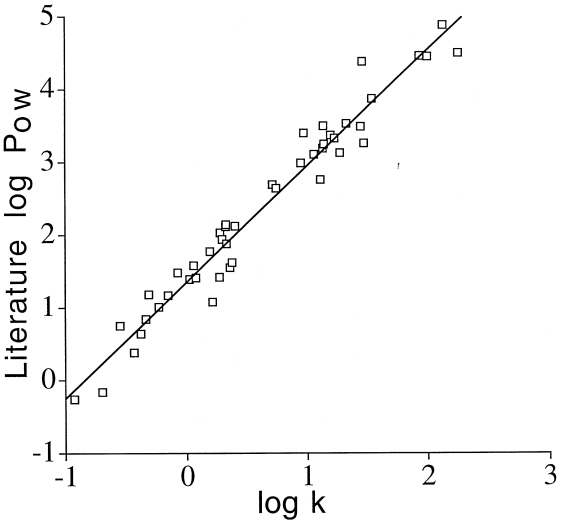


Fig. 1. Plot of literature log $P_{ow}$ against the chromatographic retention factor (log $k$) for neutral and weakly basic compounds. Retention factors were determined in the sodium dodecyl sulfate microemulsion system at pH = 10.

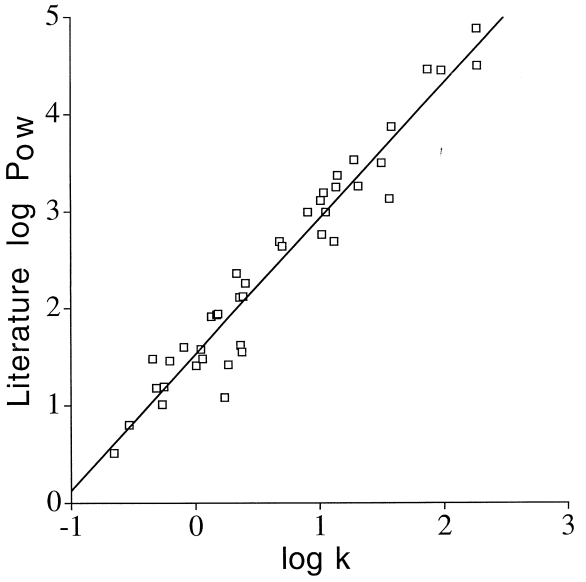


Fig. 2. Plot of literature log $P_{OW}$ against the chromatographic retention factor (log $k$) for neutral and weakly acidic compounds. Retention factors were determined in the sodium dodecyl sulfate microemulsion system at pH = 3.

microemulsion system at pH 10, Fig. 1, and 42 neutral and weakly acidic compounds run in the microemulsion system at pH 3, Fig. 2, were scrutinized. The correlation models and statistics for the fit are given in Eqs. (5) and (6) for pH = 10 and pH = 3, respectively.

$$\log P_{ow} = 1.60\ (\pm 0.05) \log k + 1.35\ (\pm 0.05) \qquad (5)$$

$$n = 45,\ \rho = 0.979,\ \mathrm{SE} = 0.27,\ F = 991$$

$$\log P_{ow} = 1.46\ (\pm 0.06) \log k + 1.46\ (\pm 0.06) \qquad (6)$$

$$n = 42,\ \rho = 0.971,\ \mathrm{SE} = 0.28,\ F = 652.$$

Both models are adequate, enabling log $P_{ow}$ to be estimated to within 0.30 log units. The models are similar but not identical because the system constant ratios differ slightly and both plots contain a few solutes with a larger than average error (>2 SE) which influence the fit.

A second set of 29 varied solutes not included in the initial evaluation set was used to validate the model for the neutral and weakly basic solutes. The compounds were selected to cover a wide range of log $P$ values (0.3–5.8). The results are summarized in Table 5 and Fig. 3. The average relative standard deviation in the estimated log $P_{ow}$ is 4.3% ($n$ = 10) and the average difference between the estimated log $P_{OW}$ and literature log $P_{OW}$ is ±0.12 log units. The slope of the plot of literature against estimated log $P_{ow}$ is 0.987 (±0.022) and intercept 0.028 (±0.063). The results are satisfactory and confirm that the sodium dodecyl microemulsion system is a useful surrogate model for the octanol–water partition system.

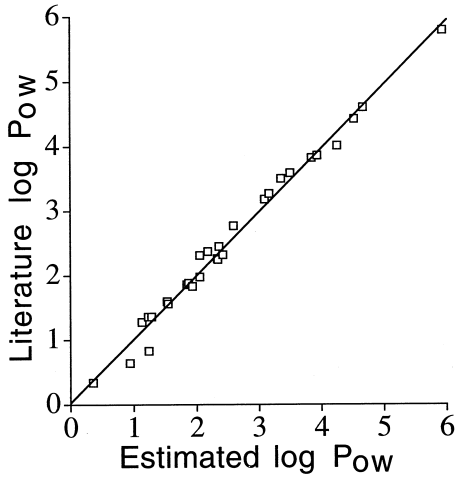


Fig. 3. Plot of literature log $P_{OW}$ against estimated log $P_{OW}$ obtained by MEEKC using the sodium dodecyl sulfate microemulsion at pH = 10.

## 4. Conclusions

Microemulsion electrokinetic chromatography (MEEKC) using a sodium dodecyl sulfate–butanol–heptane microemulsion in pH 3 and 10 buffers provides a rapid and accurate method for the estimation of log $P_{ow}$ for neutral and weakly acidic and basic compounds. The method can be fully automated using existing commercial instruments and is suitable for high sample throughput applications. The average difference in log $P_{ow}$ between literature and estimated values from MEEKC was ±0.12 log units.

Table 5
Estimated log $P_{OW}$ for the validation compounds obtained in the sodium dodecyl sulfate microemulsion system at pH = 10

| Compounds | log $P_{OW}$ experimental ($n$ = 10) | | | Literature log $P_{OW}$ | Difference (Δ) |
|---|---|---|---|---|---|
| | Mean | Standard deviation | Relative standard deviation (%) | | |
| 1-Aminonaphthalene | 2.34 | 0.131 | 5.60 | 2.25 | 0.09 |
| Benz[a]anthracene | 5.93 | 0.189 | 3.18 | 5.79 | 0.14 |
| Benzocaine | 1.84 | 0.052 | 2.81 | 1.86 | 0.02 |
| Benzophenone | 3.09 | 0.099 | 3.22 | 3.18 | 0.09 |
| Biphenyl | 4.25 | 0.071 | 1.66 | 4.01 | 0.24 |
| Butylbenzoate | 3.84 | 0.089 | 2.34 | 3.82 | 0.02 |
| Butyrophenone | 2.59 | 0.049 | 2.71 | 2.77 | 0.18 |
| 4-Chloroacetophenone | 2.42 | 0.079 | 3.26 | 2.32 | 0.10 |
| 2-Chlorobenzamide | 0.94 | 0.096 | 10.2 | 0.64 | 0.30 |
| 3-Chloropyridine | 1.13 | 0.047 | 4.22 | 1.28 | 0.15 |
| Coumarin | 1.53 | 0.128 | 8.38 | 1.60 | 0.07 |
| N,N-Diethylacetamide | 0.36 | 0.049 | 13.4 | 0.34 | 0.02 |
| N,N-Dimethylaniline | 2.05 | 0.089 | 3.36 | 2.31 | 0.26 |
| N,N-Dimethylbenzylamine | 2.05 | 0.079 | 3.89 | 1.98 | 0.07 |
| 1,3-Dimethylnaphthalene | 4.53 | 0.085 | 1.89 | 4.42 | 0.11 |
| Dimethyl phthalate | 1.54 | 0.087 | 5.65 | 1.56 | 0.02 |
| Diphenylamine | 3.35 | 0.065 | 1.94 | 3.50 | 0.15 |
| Lidocain | 2.33 | 0.059 | 2.26 | 2.26 | 0.07 |
| 2-Methoxypyridine | 1.23 | 0.063 | 5.12 | 1.36 | 0.13 |
| 2-Methylnaphthalene | 3.93 | 0.091 | 2.30 | 3.86 | 0.07 |
| Nitrobenzene | 1.93 | 0.088 | 4.56 | 1.83 | 0.10 |
| 3-Nitrotoluene | 2.36 | 0.090 | 3.82 | 2.45 | 0.09 |
| 4-Nitrotoluene | 2.18 | 0.063 | 2.90 | 2.37 | 0.19 |
| Phenyl benzoate | 3.50 | 0.100 | 2.86 | 3.59 | 0.09 |
| 1-Phenyl ethanol | 1.28 | 0.094 | 7.30 | 1.36 | 0.08 |
| Phenylurea | 1.24 | 0.051 | 4.10 | 0.83 | 0.41 |
| Pyrilamine | 3.16 | 0.096 | 3.04 | 3.27 | 0.11 |
| 1,2,4,5-Tetrachlorobenzene | 4.66 | 0.069 | 1.50 | 4.60 | 0.06 |
| 2,4,6-Trimethylpyridine | 1.87 | 0.084 | 8.40 | 1.88 | 0.01 |

## References


[1] C. Hansch, T. Fujita, J. Am. Chem. Soc. 86 (1964) 1616.
[2] A. Leo, C. Hansch, D. Elkins, Chem. Rev. 71 (1971) 525.
[3] L.-G. Danielsson, Y.-H. Zhang, Trends Anal. Chem. 15 (1996) 188.
[4] C. Hansch, P.G. Sammes, J.B. Taylor, C.A. Ramsden (Eds.), Comprehensive Medicinal Chemistry, Vol. 4, Pergamon Press, Oxford, 1990, p. 270.
[5] G.L. Biagi, A.M. Barbaro, A. Sapone, P.A. Borea, K. Varani, M. Recanatini, J. Chromatogr. A 723 (1996) 135.
[6] R. Mannhold, K. Dross, C. Sonntag, Meth. Prin. Med. Chem. 4 (1996) 141.
[7] M.H. Abraham, C.F. Poole, S.K. Poole, J. Chromatogr. A 749 (1996) 201.
[8] R. Kaliszan, Adv. Chromatogr. 33 (1993) 147.
[9] J.G. Dorsey, M.G. Khaledi, J. Chromatogr. A 656 (1993) 485.
[10] W.J. Lambert, J. Chromatogr. A 656 (1993) 469.
[11] H. van de Waterbeemed, M. Kansy, B. Wagner, H. Fischer, Meth. Prin. Med. Chem. 4 (1996) 73.
[12] K. Valko, C. Bevan, D. Reynolds, Anal. Chem. 69 (1997) 2022.
[13] K. Valko, M. Plass, C. Bevan, D. Reynolds, M.H. Abraham, J. Chromatogr. A 797 (1998) 41.
[14] C.F. Poole, S.K. Poole, A.D. Gunatilleka, Adv. Chromatogr. 40 (2000) 159.
[15] M. Hanna, V. de Biasi, B. Bond, C. Salter, A.J. Hutt, P. Camilleri, Anal. Chem. 70 (1998) 2092.
[16] M. Adlard, G. Okafo, E. Meenan, P. Camilleri, J. Chem. Soc. Chem. Commun. 21 (1995) 2241.
[17] S. Yang, J.G. Bumgarner, L.F.R. Kruk, M.G. Khaledi, J. Chromatogr. A 721 (1996) 323.
[18] M.G. Khaledi (Ed.), High Performance Capillary Electrophoresis. Theory, Techniques and Applications, Wiley, New York, 1998, p. 999.
[19] B.N. Woodrow, J.G. Dorsey, Environ. Sci. Technol. 31 (1997) 2812.

[20] Y. Ishihama, Y. Oda, K. Uchikawa, N. Asakawa, Anal. Chem. 67 (1995) 1588.
[21] M.H. Abraham, H.S. Chadha, H.J. Leo, J. Chromatogr. A 685 (1994) 203.
[22] M.H. Abraham, H.S. Chadha, R.A.E. Leitao et al., J. Chromatogr. A 766 (1997) 35.
[23] M.H. Abraham, M. Roses, C.F. Poole, S.K. Poole, J. Phys. Org. Chem. 10 (1997) 358.
[24] A. Nasal, P. Haber, R. Kaliszan, E. Forgacs, T. Cserhati, M.H. Abraham, Chromatographia 43 (1996) 484.
[25] M.H. Abraham, Chem. Soc. Rev. 22 (1993) 73.
[26] M.H. Abraham, H.S. Chadha, in: V. Pliska, B. Testa, H. van de Waterbeemed (Eds.), Lipophilicity in Drug Action and Toxicology, VCH, Weinheim, 1996, p. 311.
[27] M.H. Abraham, C.F. Poole, S.K. Poole, J. Chromatogr. A 842 (1999) 79.
[28] C.F. Poole, S.K. Poole, M.H. Abraham, J. Chromatogr. A 798 (1998) 207.
[29] S.K. Poole, C.F. Poole, Analyst 122 (1997) 267.
[30] S.K. Poole, C.F. Poole, J. High Resol. Chromatogr. 20 (1997) 174.
[31] S.K. Poole, C.F. Poole, Anal. Commun. 34 (1997) 57.
[32] M.H. Abraham, C. Treiner, M. Roses, C. Rafols, Y. Ishihama, J. Chromatogr. A 752 (1996) 243.
[33] S.J. Gluck, M.H. Benko, R.K. Hallberg, K.P. Steele, J. Chromatogr. A 744 (1996) 141.
[34] Y. Liu, D.J. Pietrzyk, J. Chromatogr. A 804 (1998) 337.
[35] M.H. Abraham, J. Phys. Org. Chem. 6 (1993) 660.
[36] M.H. Abraham, J. Andonian-Haftvan, G.S. Whiting, A. Leo, R.S. Taft, J. Chem. Soc. Perkin Trans. 2 (1994) 1777.
[37] M.H. Abraham, M. Roses, J. Phys. Org. Chem. 7 (1994) 672.
[38] S.K. Poole, C.F. Poole, J. Chromatogr. A 845 (1999) 381.
[39] C. Hansch, A. Leo, D. Hoekman, Exploring QSAR: Hydrophobic, Electronic and Steric Constraints, Vols. 1 and 2, American Chemical Society, Washington, DC, 1995.