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Application of a modified linear solvation energy relationship (LSER) model to retention on a butylimidazolium-based column for high performance liquid chromatography

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ABSTRACT

Previously, a new HPLC stationary phase based on *n*-butylimidazolium bromide was investigated using a linear solvation energy relationship (LSER) to systematically evaluate the intermolecular interactions between 32 test solutes and the stationary phase. The results and further comparisons with conventional reversed phase systems revealed that retention properties are similar to phenyl phases in both methanol/water and acetonitrile/water mixtures. In this work, the LSER model is extended by including the degree of ionization molecular descriptor, *D*, which takes into account the pK_a of ionizable analytes and the pH of the mobile phase. The *D* molecular descriptor has been further divided into D^+ and D^- components that separately account for the ionization of basic and acidic solutes, respectively. This is the first study where the ionization terms for weakly acidic solutes and weakly basic solutes have been separated. LSER results obtained with the expanded solute set with and without the inclusion of the D^+ and D^- solute descriptors were compared. The improved correlation and standard error obtained for the expanded test set in the presence and absence of the D^+ and D^- descriptors (R^2: 0.987 vs 0.846; SE: 0.051 vs 0.163 for 60% MeOH) support inclusion of these additional terms. Further, the coefficients obtained from the multiple linear regression for the expanded test set with the D^+ and D^- descriptors were more consistent with the coefficients obtained when the test set included just neutral analytes. In addition, the expanded LSER model did a better job of predicting elution order for the ionizable analytes. This work provides further supporting evidence for the multimodal nature of the butylimidazolium stationary phase.

1. Introduction

The linear solvation energy relationship (LSER) model has been shown to be quite successful in accounting for retention of neutral compounds on a variety of reversed phase liquid chromatographic columns (RPLC) [1–19]. It would also be useful, however, if this model could be used in the analysis of ionizable compounds. Indeed, the retention of ionizable compounds in RPLC is different from that of neutral compounds because the retention of neutrals is independent of the mobile phase pH at any fixed composition. In contrast, the retention of ionizable compounds is highly pH-dependent because the equilibrium distribution of the acidic and basic forms is affected by the pH [20].

Several groups have studied the fundamental retention behavior of ionizable analytes in reversed-phase chromatography [21–24]. Indeed, recent investigations by Barbosa [25–28], Sykora [29,30], Heinisch [31] and Roses [32–36] focused on the impact of the addition of organic modifiers on the pH of the mobile phase and the ionization of these compounds. In general, it has been reported that the addition of methanol, for instance, results in an increased pH of an acidic mobile phase and pK_a of weakly acidic compounds simultaneously, but a decreased pK_a of weakly basic compounds. In the absence of ion-pairing agents, reduced retention is seen in conventional reversed phase systems for all ionized compounds when compared to their neutral analogues [37].

Several attempts have been made to modify the LSER model so that it can accommodate ionizable analytes resulting in the development of two possible descriptors [33,38,39]. The *P* solute descriptor describes the effective acid dissociation constant for a given mobile phase composition and is calculated using Eq. (1) [38]:

$$P = \frac{14 - \mathrm{p}K_a}{10} \quad (1)$$

The modified LSER model including the *P* solute descriptor was found to be viable for analyte sets with both neutral and ionizable compounds [38]. It was noted, however, that predictions of this

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model may be poor because the mobile phase pH is not taken into account [39]. Another solute descriptor, D, describes the degree of ionization of the solute at the pH of the mobile phase [33]. The D solute descriptor is calculated using Eq. (2) [33]:

$$D = \frac{[X^-]}{[HX] + [X^-]} = \frac{10^{pH-pK_a}}{(1 + 10^{pH-pK_a})} \quad (2)$$

This modified LSER model, including this descriptor, given by Eq. (3):

$$\log k = c + eE + sS + aA + bB + vV + dD \quad (3)$$

was found to work well for retention of solute sets containing both neutral and acidic compounds at all pH values under reversed phase conditions [38].

The non-pH corrected LSER model has been used to characterize retention for neutral analytes on a novel butylimidazolium-based column [40,41]. In contrast to reversed phase systems, where the ionization of both acids and bases leads to retention losses, ionizable analytes would be expected to exhibit bimodal behavior on the butylimidazolium stationary phase. For instance, the ionization of weakly acidic analytes should lead to an increase in retention whereas ionization of weakly basic compounds (e.g., amines) should lead to a reduction in retention. The increased retention observed for weakly acidic analytes could be attributed to attractive electrostatic interactions between the immobilized cation and an anionic analyte while the reduction in retention for weakly basic analytes could be attributed to the electrostatic repulsion between a cation and the immobilized imidazolium cation [42].

Retention data was obtained using 60% and 70% MeOH for a test set that included weakly acidic (e.g., phenols) and weakly basic (e.g., pyridine, aniline, 2-chloroaniline) solutes. These mobile phase compositions were selected because of the excellent correlation between experimental and predicted retention for the neutral probe solutes reported previously [41]. In addition, at higher organic mobile phase compositions, the low analyte retention relative to the void volume leads to larger relative error in the measurements. At lower organic mobile phase compositions, adventitious ions in the mobile phase may exchange with the stationary phase counter-ion, thereby altering the analyte:stationary phase interactions [41]. Hence, 60% and 70% MeOH represent a reasonable compromise. Under these conditions, weakly acidic (e.g., nitrophenols with pK_a = 7.2–8.4 in water) [43] and weakly basic compounds (e.g., pyridine with pK_a = 5.2 in water) [43] have been well studied [31,36] and could be considered as good weakly acidic and basic representative compounds. The acidic and basic solutes were accounted for in the LSER model using the D^- and D^+ molecular descriptors, respectively. The chromatographic data was analyzed using both the original and the D^+/D^- modified LSER models. The results obtained with both models are discussed and compared to assess the suitability of the extended model to chromatographic data obtained on the butylimidazolium-based column. This is the first time that the degree of ionization molecular descriptor has been split into separate terms to simultaneously account for the ionization of acidic and basic solutes.

2. Experimental

2.1. Materials

The previously used training set [40,41], consisting of 28 probe solutes, was supplemented with weakly acidic and weakly basic solutes in these studies. The probe solutes and their molecular solute descriptors [44,45] are shown in Table 1. All chemicals, including the probe solutes and mobile-phase components (HPLC-grade water and methanol), were purchased from the Aldrich Chemical Company (Milwaukee, WI), the Fisher Scientific Chemical Company (Fair Lawn, NJ) or Pharmco Products Inc. (Brookfield, CT) and used without further purification.

Table 1
The probe solutes and their solute descriptors.

Probe solute	Descriptors				
	E	*S*	*A*	*B*	*V*
1. Benzene	0.610	0.52	0.00	0.14	0.716
2. Naphthalene	1.340	0.92	0.00	0.20	1.085
3. Biphenyl	1.360	0.99	0.00	0.22	1.324
4. Anthracene	2.290	1.34	0.00	0.26	1.454
5. Toluene	0.601	0.52	0.00	0.14	0.857
6. o-Xylene	0.663	0.56	0.00	0.16	0.998
7. Mesitylene	0.649	0.52	0.00	0.19	1.139
8. Ethylbenzene	0.613	0.51	0.00	0.15	0.998
9. Propylbenzene	0.604	0.50	0.00	0.15	1.139
10. n-Butylbenzene	0.600	0.51	0.00	0.15	1.280
11. tert-Butylbenzene	0.619	0.49	0.00	0.16	1.280
12. Fluorobenzene	0.477	0.57	0.00	0.10	0.734
13. Chlorobenzene	0.718	0.65	0.00	0.07	0.839
14. Iodobenzene	1.188	0.82	0.00	0.12	0.975
15. 1,2-Dichlorobenzene	0.872	0.78	0.00	0.04	0.961
16. Phenol	0.805	0.89	0.60	0.30	0.775
17. Benzyl alcohol	0.803	0.87	0.33	0.56	0.916
18. 2-Phenylethanol	0.811	0.91	0.30	0.64	1.057
19. p-Cresol	0.820	0.87	0.57	0.31	0.916
20. p-Chlorophenol	0.915	1.08	0.67	0.20	0.898
21. Nitrobenzene	0.871	1.11	0.00	0.28	0.891
22. Benzonitrile	0.742	1.11	0.00	0.33	0.871
23. Benzaldehyde	0.820	1.00	0.00	0.39	0.873
24. Aniline	0.955	0.96	0.26	0.50	0.816
25. Acetophenone	0.818	1.01	0.00	0.48	1.014
26. 2-Chloroaniline	1.033	0.92	0.25	0.40	0.939
27. Anisole	0.708	0.75	0.00	0.29	0.916
28. n-Butylbenzoate	0.688	0.80	0.00	0.46	1.495
29. Pyridine	0.631	0.84	0.00	0.47	0.675
30. p-Nitrophenol	1.070	1.72	0.82	0.26	0.949
31. o-Nitrophenol	1.015	1.05	0.05	0.37	0.949
32. m-Nitrophenol	1.050	1.57	0.79	0.23	0.949

2.2. Methods

All HPLC experiments with both the training set and ionizable compounds were performed using Shimadzu (Columbia, MD) LC-10AT pumps at room temperature. Detection for the training set was accomplished using a SPD-10A UV detector set at 254 nm or 310 nm. The butylimidazolium column (Nucleosil silica, 100 Å, 5 μm, 350 m^2/g) was 250 mm × 4.6 mm I.D. was prepared in house as previously discussed [41].

All probe solutes were dissolved (0.004–0.5 mg/mL) in methanol (neutral solutes) or methanol/water mixtures (60% and 70% methanol for ionizable solutes). A 20 μL aliquot of the sample was injected using a Rheodyne injection valve (Cotati, CA) with all experiments performed in triplicate. Sample retention was found to have a standard deviation of 0.022 min or less. The flow rate was 0.8 mL/min. The Accumet model XL150 pH meter (Fisher Scientific, Fair Lawn, NJ) was standardized at room temperature with aqueous buffers of pH 4.0, 7.0 and 10.0. The pH measurements for the 60% and 70% methanol/water mixtures were made in triplicate to ±0.05 pH units and had apparent pH values of 7.45 and 8.20, respectively.

The pK_a values for the ionizable compounds in water and calculated in methanol–water according to literature [45,46] are summarized in Table 2. It should be noted that for acidic compounds, the D descriptor is calculated by Eq. (2); for basic compounds, it is calculated using Eq. (4):

$$D = \frac{[BH^+]}{([B] + [BH^+])} = \frac{10^{pK_a-pH}}{(1 + 10^{pK_a-pH})} \quad (4)$$

Table 2
The pK and D solute descriptor values for the ionizable compounds in water and a 60% or 70% methanol/water solution and their reference values in water.

Analytes	pK_{aq}	pK_{MeOH}	$D_{aq,\ pH\ 6.5}$	p$K_{60\%\ MeOH}$[a]	$D_{60\%\ MeOH}$	p$K_{70\%\ MeOH}$[a]	$D_{70\%\ MeOH}$
Pyridine	5.29	5.43	9.94E−01	4.15	4.97E−04	4.00	6.29E−05
Phenol	9.97	14.32	3.40E−04	11.00	2.84E−04	11.22	3.40E−04
p-Cresol	10.26	14.54	1.75E−04	11.26	1.56E−04	11.49	5.17E−04
p-Chlorophenol	9.38	13.59	1.32E−03	10.39	1.14E−03	10.63	3.72E−03
Aniline	4.59	6.04	9.99E−01	4.19	5.55E−04	4.09	7.68E−05
2-Chloroaniline	2.66	3.49	1.00E0	2.33	7.57E−06	2.22	1.06E−06
p-Nitrophenol	7.15	11.24	1.73E−01	7.81	3.05E−01	8.03	5.95E−01
o-Nitrophenol	7.24	11.52	1.60E−01	8.04	2.04E−01	8.27	4.61E−01
m-Nitrophenol	8.43	12.40	1.36E−02	9.11	2.15E−02	9.32	7.01E−02

[a] pK reference data and pK estimates based on Refs. [45,46].

The D solute descriptors for pyridine and aniline were also determined spectrophotometrically. All spectroscopy experiments were done using a Varian Cary 50 Bio UV-Visible Spectrometer (Santa Clara, CA). Standard solutions of the analytes (3 mM) were prepared in 60% MeOH and 70% MeOH. The absorbance spectra were obtained for these standard solutions and for aliquots of the standard solution to which either NaOH or HCl had been added. Absorbance at an appropriate wavelength (e.g., λ = 256 nm for pyridine) was used to estimate the extent of ionization in the standard solution.

The void volume for the butylimidazolium-based column was determined by measuring the difference in column weights when the column was filled with methylene chloride or with hexane. Chromatographic retention data was acquired with a Chrom&Spec Chromatography Data System (Ampersand International, Inc., Beachwood, OH). Multiple linear regression analysis and statistical tests of the chromatographic data were performed on a PC using Excel.

3. Results and discussion

The appropriateness of the solute set selected and the modified LSER model was evaluated in several ways. These methods include an examination of the covariance matrix of the molecular solute descriptors, plots of predicted log k vs experimental log, generated LSER model statistics and whether the predicted elution order of ionizable solutes is actually observed. Favorable results in these areas would indicate that the LSER model modified with the D solute descriptor does a good job of describing the retention of the solute set.

3.1. Covariance of selected LSER molecular solute descriptors

The molecular solute descriptors, shown in Table 1, were used to construct a covariance matrix, shown in Table 3, to evaluate the suitability of the solute test set for use with the LSER model. In contrast to the other molecular descriptors, which are independent of mobile phase composition, the D molecular descriptors are sensitive to mobile phase conditions. Hence, terms are included in the table for both compositions used in this study. Ideally, the correlation coefficients for the molecular solute descriptors will be low. When there is high covariance between parameters, the uncertainties in the LSER coefficients are inflated because the model does not know how to distribute variance in the data. The correlation matrix for the individual solute descriptors indicated that there is a moderate correlation between E and S (0.607). A correlation between these terms has been previously noted for both aliphatic and aromatic solute sets and can be explained by the fact that both terms reflect a contribution to retention due to the polarizability of the solute [47]. There is also a moderate correlation between A and S (0.608) that can be attributed to the ion-induced dipole interaction capabilities of the solute. The generally low correlation coefficients imply that this solute test set was appropriate to be used with the LSER model.

3.2. Evaluation of LSER model quality

Fig. 1 shows the LSER predicted log k vs experimental log k results for the butylimidazolium column when chromatographic data for weakly acidic and weakly basic solutes are included for (a) 60% and (b) 70% methanol. As can be seen in Fig. 1, inclusion of retention data for the ionizable analytes (the circled outliers) on the butylimidazolium phase and application of the LSER model without accounting for ionization seriously degrades the correlation between predicted and experimental results.

The D solute descriptor has mainly been used previously to describe the ionization of acidic compounds [26–28,35,39]. Few studies have been reported in which results for basic solutes [30] or both basic and acidic solutes together [33] have been examined. Therefore, the LSER modified with the D solute descriptor was applied first to the test set which included neutral and weakly acidic (nitrophenols, cresol and p-chlorophenol) solutes and then to the test set including neutral and weakly basic (pyridine, aniline and 2-chloroaniline) solutes in order to assess whether the D solute descriptor can adequately describe both sets of compounds in this separation system. Fig. 2 shows the LSER predicted log k vs experimental log k for the neutral and acidic compounds while Fig. 3 shows the LSER predicted log k vs experimental log k for the neutral and basic compounds, both with (a) 60% and (b) 70% methanol.

The linearity of the plots in Figs. 2 and 3 imply that the LSER model modified with the D solute descriptor adequately describes the retention of both neutral and acidic solutes or neutral and

Table 3
Correlation coefficient matrix for solute descriptors.

	E	S	A	B	V	D_{60}^-/D_{70}^-	D_{60}^+/D_{70}^+
E	1	0.607	0.115	0.059	0.401	−0.049/−0.044	0.141/0.143
S		1	0.607	0.340	0.000	0.034/0.036	0.491/0.490
A			1	0.203	−0.226	−0.001/−0.001	0.347/0.344
B				1	−0.026	0.372/0.371	0.063/0.065
V					1	−0.316/−0.311	−0.055/−0.056
D_{60}^-/D_{70}^-						1/1	−0.069/−0.069
D_{60}^+/D_{70}^+							1/1

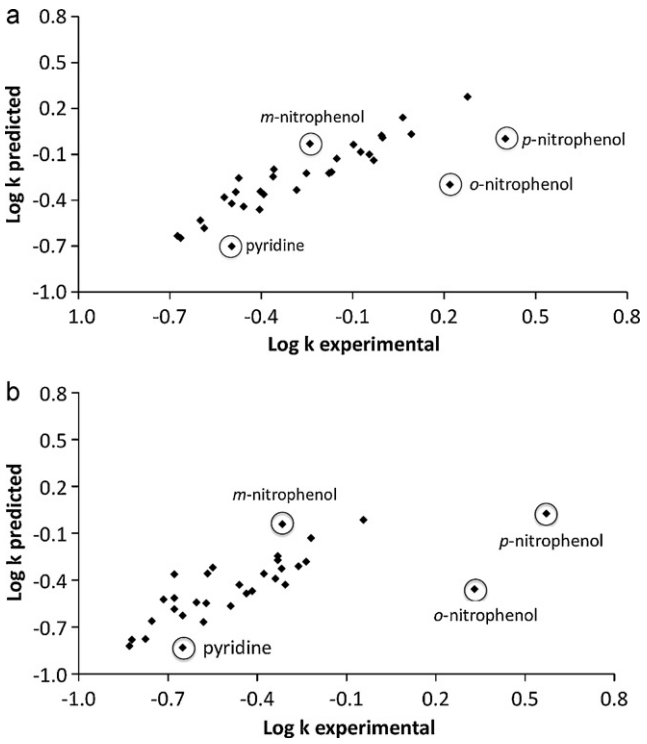

Fig. 1. Plot of $\log k_{predicted}$ vs $\log k_{experimental}$ using the original LSER model at (a) 60% MeOH or (b) 70% MeOH mobile phase composition.

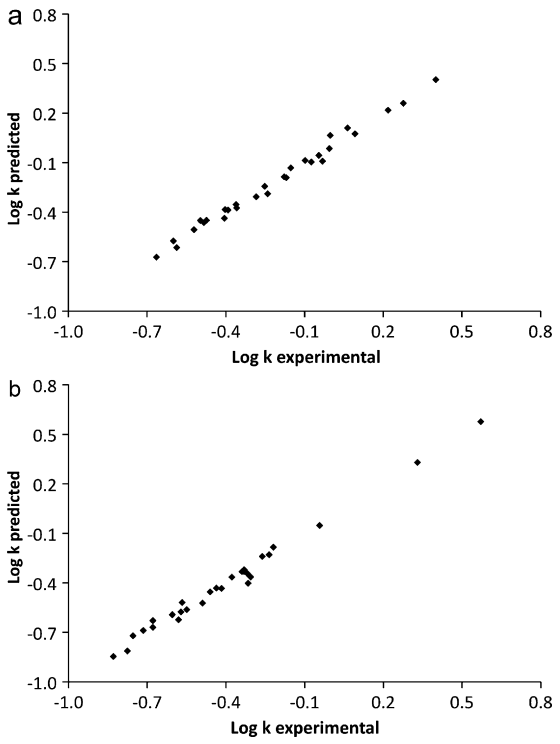

Fig. 2. Plot of $\log k_{predicted}$ vs $\log k_{experimental}$ for acidic and neutral solutes using the *D*-modified LSER model described in Eq. (3) at (a) 60% MeOH or (b) 70% MeOH mobile phase composition.

basic solutes. Further, the addition of the *D* solute descriptor significantly improves the regression statistics for both the neutral and acidic solutes and the neutral and basic solutes as shown in Tables 4 and 5. The effect is more prominent with the acidic solutes, perhaps because their *D* solute descriptors cover a larger dynamic range than do the basic solutes. However, when the LSER model is used for all of the neutral, acidic and basic solutes at once, pyridine becomes a significant outlier as shown in Fig. 4. Because the modified LSER model adequately describes retention for acidic and basic solutes when they are analyzed separately but not when they are together, it is necessary to account for solutes that are acidic and those that are basic separately in the model. Abraham and Acree encountered a similar issue in their study of retention for cations and anions [48,49]. However their solute set consisted primarily of the conjugate acids and bases of strong acids and bases.

When weak acids and bases were included, they were assumed to be fully ionized like their strong counterparts, which is not appropriate under the experimental conditions used in the current study. Further, it has been noted by Krygowski and Fawcett that complementary interactions must be accounted for separately because the species capable of these types of interactions act independently of one another [50]. Hence, the complementary interactions of deprotonated acids and protonated bases might be better accounted for separately. For clarity, the *D* solute descriptor and *d* coefficient for acidic solutes will be denoted D^- and d^-, respectively whereas D^+ and d^+ will denote basic solutes. Thus, the modified LSER model becomes:

$$\log k = c + eE + sS + aA + bB + vV + d^+D^+ + d^-D^- \quad (5)$$

Fig. 5 shows a plot of all acidic, basic and neutral solutes using the modified LSER model that separates the terms accounting for acidic and basic solutes. The linearity of this plot indicates that this modified LSER model does a much better job of describing the retention

Table 4
LSER coefficients and predicted vs experimental regression statistics for neutral and acidic solutes using the original LSER or *D* modified LSER model described in Eq. (3).

	60% MeOH			70% MeOH		
	Without correction neutrals only[a]	Without correction[b]	With *D* descriptor[b]	Without correction neutrals only[a]	Without correction[b]	With *D* descriptor[b]
c	−0.96 ± 0.04	−1.19 ± 0.19	−1.02 ± 0.04	−1.00 ± 0.04	−1.33 ± 0.28	−1.08 ± 0.04
e	0.11 ± 0.04	−0.11 ± 0.15	0.06 ± 0.03	0.12 ± 0.03	−0.20 ± 0.21	0.05 ± 0.03
s	−0.06 ± 0.07	0.47 ± 0.19	0.04 ± 0.04	−0.06 ± 0.05	0.72 ± 0.27	0.09 ± 0.05
a	−0.13 ± 0.03	−0.13 ± 0.17	−0.10 ± 0.03	−0.07 ± 0.09	−0.11 ± 0.24	−0.06 ± 0.04
b	−0.88 ± 0.09	−1.15 ± 0.25	−0.98 ± 0.05	−0.79 ± 0.08	−1.16 ± 0.35	−0.92 ± 0.05
v	0.89 ± 0.04	0.97 ± 0.19	0.93 ± 0.04	0.66 ± 0.03	0.78 ± 0.28	0.72 ± 0.04
d			4.14 ± 0.17			2.40 ± 0.08
R	0.995	0.836	0.995	0.995	0.707	0.995
SE	0.029	0.167	0.032	0.022	0.240	0.036
F	332	11	350	311	5	342

R, overall correlation coefficient; SE, standard error in estimate; *F*, *f*-statistic.
[a] Number of solutes = 24.
[b] Number of solutes = 29.

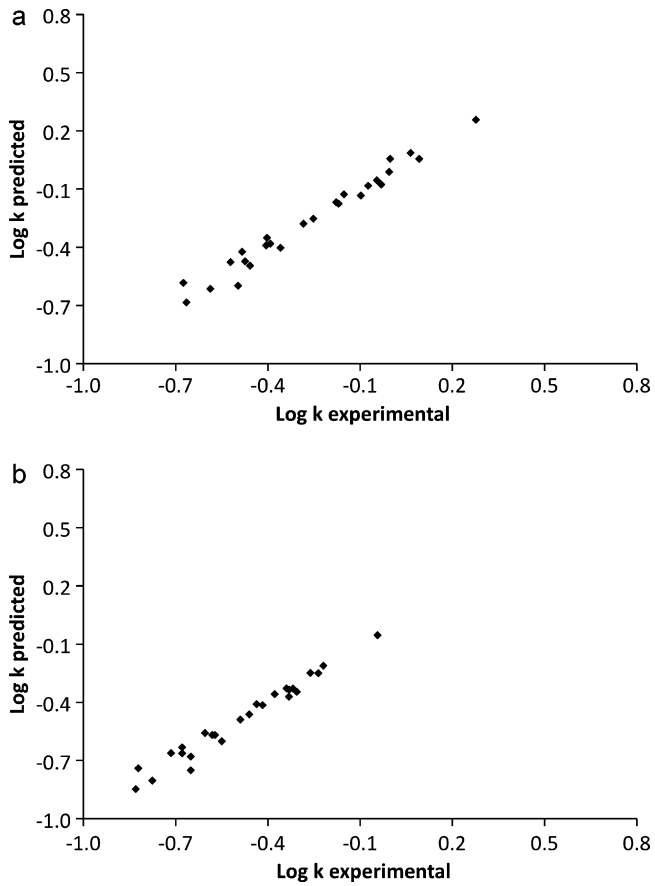

Fig. 3. Plot of log $k_{predicted}$ vs log $k_{experimental}$ for basic and neutral solutes using the *D*-modified LSER model described by Eq. (3) at (a) 60% MeOH or (b) 70% MeOH mobile phase composition.

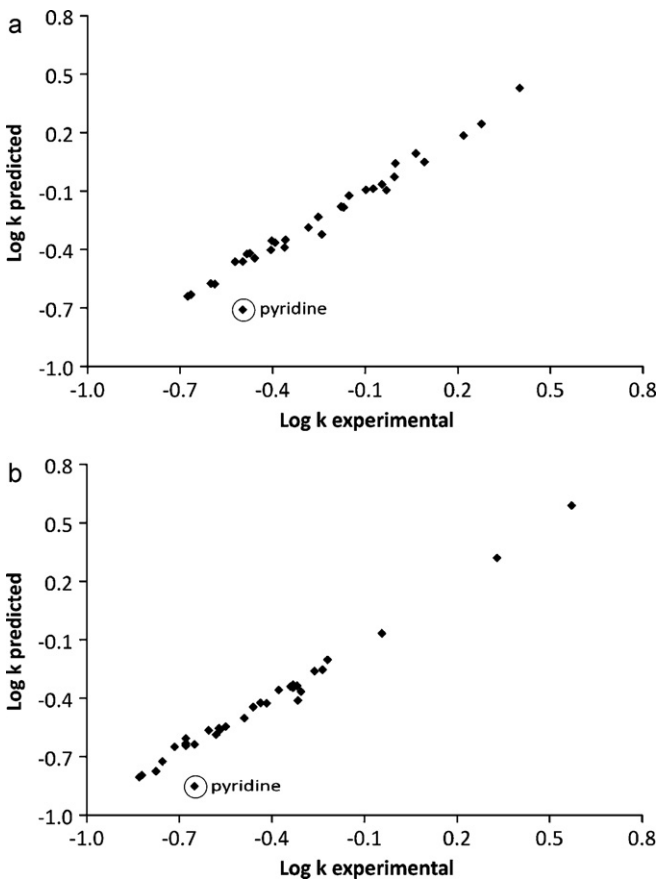

Fig. 4. Plot of log $k_{predicted}$ vs log $k_{experimental}$ for acidic, basic and neutral solutes using the *D*-modified LSER model described in Eq. (3) at (a) 60% MeOH or (b) 70% MeOH mobile phase composition.

of all the solutes examined than a single *D* descriptor. To the best of our knowledge, this is the first time the *D* solute descriptor has been split into two terms.

It should be noted that aniline and pyridine continue to lie slightly away from the best-fit line. Poor fits can be the result of having solutes whose descriptors lack wide variation [47]. However, it should be noted that only three basic compounds have been examined here, compared to six acidic compounds. Further, the D^- solute descriptors cover four orders of magnitude while the D^+ solute descriptors cover only two orders of magnitude.

The possibility that the various models for estimating the p*K* on which the D^+ solute descriptors are based often yield slightly different values was also considered. A range of pK_a values for both the acidic and basic solutes were used to calculate the D^+/D^- molecular descriptors [46]. These values were then substituted into the extended LSER model. The basic solutes proved much more sensitive to pK_a values than the phenols. Hence, the D^+ solute descriptors for pyridine and aniline were also determined spectroscopically. Table 6 shows a comparison of the *D* solute descriptors determined by the literature model and spectroscopic methods. When the *D* solute descriptors determined spectroscopically were used

Table 5
LSER coefficients and predicted vs experimental regression statistics for neutral and acidic solutes using the original LSER or *D* modified LSER model described in Eq. (3).

	60% MeOH			70% MeOH		
	Without correction neutrals only[a]	Without correction[b]	With *D* descriptor[b]	Without correction neutrals only[a]	Without correction[b]	With *D* descriptor[b]
c	−0.96 ± 0.04	−0.82 ± 0.07	−0.88 ± 0.07	−1.00 ± 0.04	−0.87 ± 0.06	−0.92 ± 0.06
e	0.11 ± 0.04	0.20 ± 0.06	0.17 ± 0.06	0.12 ± 0.03	0.21 ± 0.06	0.18 ± 0.05
s	−0.06 ± 0.07	−0.24 ± 0.11	−0.18 ± 0.10	−0.06 ± 0.05	−0.23 ± 0.10	−0.17 ± 0.09
a	−0.13 ± 0.03	−0.54 ± 0.16	−0.46 ± 0.15	−0.07 ± 0.09	−0.47 ± 0.14	−0.41 ± 0.13
b	−0.88 ± 0.09	−0.51 ± 0.13	−0.64 ± 0.14	−0.79 ± 0.08	−0.55 ± 0.12	−0.55 ± 0.12
v	0.89 ± 0.04	0.73 ± 0.06	0.80 ± 0.06	0.66 ± 0.03	0.52 ± 0.06	0.57 ± 0.06
d			181 ± 81			1191 ± 541
R	0.995	0.983	0.987	0.995	0.979	0.984
SE	0.029	0.051	0.047	0.022	0.046	0.042
F	332	118	119	311	96	96

R, overall correlation coefficient; SE, standard error in estimate; *F*, *f*-statistic
[a] Number of solutes = 24.
[b] Number of solutes = 26.

Table 6
Comparison of the *D* solute descriptors determined using a literature model or spectroscopy.

	60% MeOH		70% MeOH	
	Literature model[a]	Spectroscopy	Literature model[a]	Spectroscopy
Pyridine	4.97E−04	1.94E−04	6.29E−05	5.38E−05
Aniline	5.55E−04	1.50E−05	7.68E−05	1.39E−05

[a] Refs. [45,46].

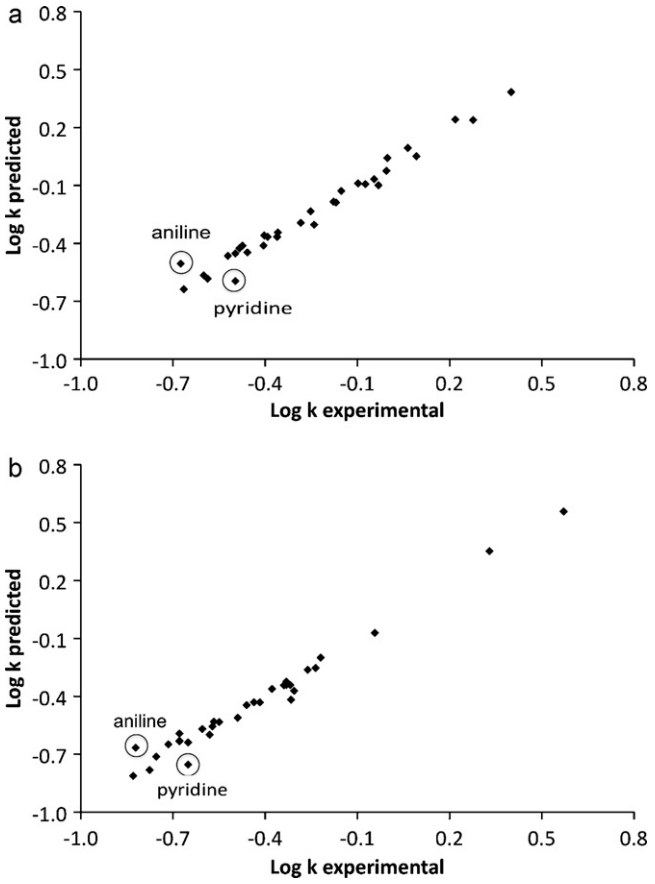

Fig. 5. Plot of log $k_{predicted}$ vs log $k_{experimental}$ for acidic, basic and neutral solutes using the D^+ and D^--modified LSER model described in Eq. (5) at (a) 60% MeOH or (b) 70% MeOH mobile phase composition.

with the modified LSER model, the plot of predicted log *k* vs experimental log *k* has no outliers. This is shown in Fig. 6 where all acidic, basic and neutral solutes are adequately described by the modified LSER model. It should also be noted that when the spectroscopically determined *D* coefficient for aniline was used in the extended model incorporating a single *D* term, pyridine was still a significant outlier. Therefore, the spectroscopically determined *D* solute descriptors for pyridine and aniline were used while the *D* solute descriptors employed for the remaining ionizable solutes were determined using a model reported in the literature [45,46].

3.3. Evaluation of LSER coefficients and regression statistics

The LSER coefficients and regression statistics for predicted log *k* vs experimental log *k* for the subset of original 28 neutral analytes and the complete set including the weakly acidic and weakly basic solutes using the original LSER model and D^+ and D^--modified LSER model, are shown in Table 7 for 60% and 70% methanol. As expected, the *v* coefficient for the neutral compounds is large and positive while the others are smaller and positive or negative. One of the most striking features of the data including the additional analytes is the significant improvement in correlation between calculated and experimental results when the D^+ and D^- terms are used to correct the original LSER model. The correlation between the calculated and the experimental results increased from 0.85 to 0.99 for 60% methanol and from 0.73 to 0.99 for 70% methanol.

In the reversed phase system reported previously [38], the *d* coefficient was negative, denoting a repulsive interaction with the stationary phase. For a reversed phase system, this seems reasonable as a charged species would not partition into a neutral stationary phase except as an ion-pair. The significant difference between the butylimidazolium system and the reversed phase system provides supporting evidence for the incorporation of this imidazolium cation in the stationary phase. Further, it indicates that, at least for the phenol-based solutes, this molecular descriptor successfully accounts for the attractive interactions between the analytes and the butylimidazolium-based stationary phase.

It is also interesting to note that both the d^+ and d^- coefficients derived from the modified LSER model are positive values for the butylimidazolium system, denoting an overall attractive interaction with the stationary phase. It makes chemical sense for the

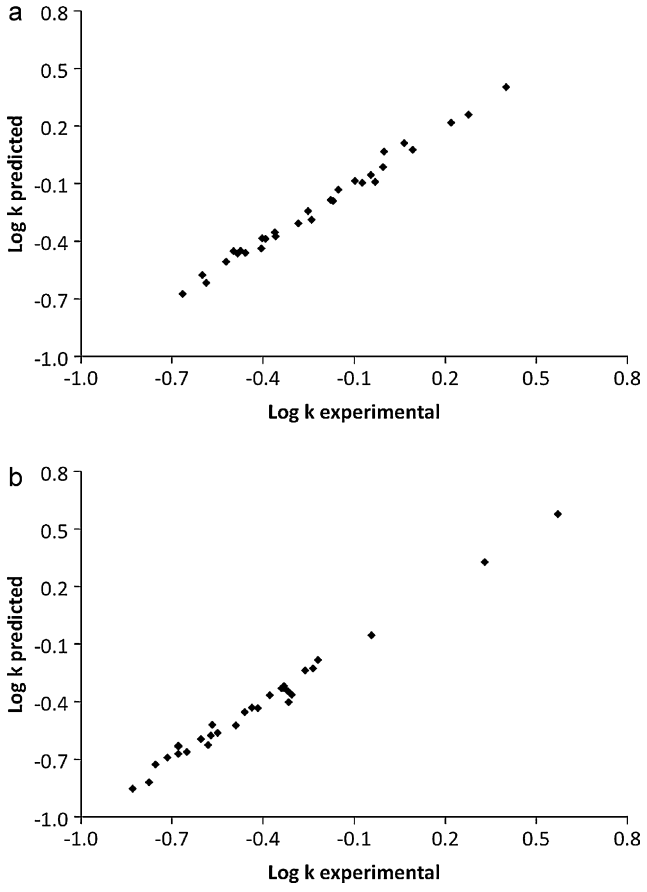

Fig. 6. Plot of log $k_{predicted}$ vs log $k_{experimental}$ for acidic, basic and neutral solutes using the D^+/D^- modified LSER model described by Eq. (5) where D^+ for aniline and pyridine were estimated using spectroscopy at (a) 60% MeOH or (b) 70% MeOH mobile phase composition.

Table 7
LSER coefficients and predicted vs experimental regression statistics for the original LSER or D^+/D^- modified LSER model described in equation 5 using the D^+ solute descriptors determined spectroscopically for pyridine and aniline.

	60% MeOH			70% MeOH		
	Without correction neutrals only[a]	Without correction[b]	With D^+/D^- descriptor[b]	Without correction neutrals only[a]	Without correction[b]	With D^+/D^- descriptor[b]
c	−0.96 ± 0.04	−1.13 ± 0.18	−1.02 ± 0.04	−1.02 ± 0.04	−1.28 ± 0.25	−1.09 ± 0.04
e	0.11 ± 0.04	−0.12 ± 0.14	0.06 ± 0.03	0.06 ± 0.03	−0.20 ± 0.19	0.04 ± 0.03
s	−0.06 ± 0.07	0.49 ± 0.18	0.04 ± 0.04	−0.06 ± 0.05	0.73 ± 0.25	0.10 ± 0.04
a	−0.13 ± 0.03	−0.18 ± 0.16	−0.10 ± 0.04	−0.07 ± 0.09	−0.15 ± 0.22	−0.07 ± 0.03
b	−0.88 ± 0.09	−1.08 ± 0.21	−0.98 ± 0.04	−0.79 ± 0.08	−1.10 ± 0.30	−0.93 ± 0.05
v	0.89 ± 0.04	0.90 ± 0.17	0.93 ± 0.03	0.66 ± 0.03	0.71 ± 0.24	0.73 ± 0.04
d^+			1491 ± 179			5047 ± 759
d^-			4.14 ± 0.16			2.40 ± 0.11
R	0.995	0.846	0.995	0.995	0.728	0.995
SE	0.029	0.163	0.1630	0.2	0.229	0.036
F	332	13	374	311	6	326

R, overall correlation coefficient; SE, standard error in estimate; F, f-statistic.
[a] Number of solutes = 24.
[b] Number of solutes = 32.

d^- coefficient to be positive because negatively charged ions are expected to be attracted to the positively charged imidazolium moiety. One might expect the d^+ coefficient to be negative, because of repulsion from the imidazolium moiety. The fact that the d^+ coefficient is also positive implies that these solutes are interacting either with the imidazolium counter ion or with residual silanols.

The trends in the values for the d^+ and d^- coefficients also make chemical sense. As the percent methanol in the mobile phase increases, the pK_a of acidic solutes increases while that of basic solutes decreases [51]. As a result, the D solute descriptor values increase (meaning more ionized) for acidic solutes and decrease (meaning less ionized) for basic solutes as the percent methanol in the mobile phase increases. It follows, therefore, that the d^- coefficient increases while the d^+ coefficient decreases with increasing percent methanol in the mobile phase. The overall result is that the value of the d^-D^- term increases with methanol concentration while the value of the d^+D^+ term decreases.

Comparison of the coefficients in Table 7 obtained for the data set containing only neutral analytes and those coefficients obtained for the data set containing the additional ionizable analytes without accounting for electrostatic interactions reveals that the coefficients seem to fall into two groups at both methanol concentrations. The a, b and v coefficients are not significantly affected by the incorporation of the d^+D^+ and d^-D^- terms. For example, at 60% methanol, the dispersion term, v, has a value of 0.89 when the data set contains only neutrals and 0.90 without correction for analyte ionization. Previous reversed phase LSER studies on the butylimidazolium-based column yielded a and b coefficients with negative signs and a v coefficient with a positive sign [40,41]. Therefore, it seems reasonable that these coefficients retain those signs here, although the magnitude of the coefficient may have changed slightly. Further, the uncertainties associated with the coefficients introduced by the inclusion of the ionizable solutes are significantly reduced when the D descriptors are included.

The second group consists of the e and s coefficients, which are moderately affected by the incorporation of the d^+D^+ and d^-D^- terms. For instance, the −0.12 value obtained for the e coefficient (at 60% methanol, for example) without the analyte ionization correction is not reasonable because previous reversed phase LSER studies on the butylimidazolium-based column yielded a positive sign [40,41]. This implies that some ion–dipole type interactions are incorporated into this term in the absence of the d^+D^+ and d^-D^-

Table 8
Products of solute descriptors and LSER coefficients for acidic and basic solutes accounting for ionization with the d^+D^+ and d^-D^- terms at 60% methanol.

Solute	eE	sS	aA	bB	vV	c	d^+D^+	d^-D^-	$\log k_p$[a]	$\log k_e$[b]	$\log k_e - \log k_p$
Original											
Pyridine	−0.074	0.410	0.60	−0.508	0.607	−1.134			−0.700	−0.497	0.203
Phenol	−0.094	0.433	−0.108	−0.324	0.697	−1.134			−0.531	−0.600	−0.069
p-Cresol	−0.096	0.424	−0.103	−0.335	0.823	−1.134			−0.421	−0.497	−0.076
p-Chlorophenol	−0.107	0.525	−0.121	−0.216	0.807	−1.134			−0.245	−0.361	−0.116
Aniline	−0.112	0.467	−0.047	−0.540	0.734	−1.134			−0.632	−0.675	−0.043
2-Chloroanaline	−0.121	0.448	−0.045	−0.432	0.844	−1.134			−0.440	−0.458	−0.018
p-Nitrophenol	−0.125	0.837	−0.148	−0.281	0.853	−1.134			0.003	0.401	0.398
o-Nitrophenol	−0.119	0.511	−0.009	−0.400	0.853	−1.134			0.219	−0.297	0.516
m-Nitrophenol	−0.123	0.764	−0.142	−0.249	0.853	−1.134			−0.241	−0.030	−0.210
Modified											
Pyridine	0.036	0.033	0.0	−0.462	0.627	−1.021	0.289	0.000	−0.497	−0.497	0.000
Phenol	0.046	0.035	−0.062	−0.295	0.720	−1.021	0.0000	0.001	−0.576	−0.600	−0.024
p-Cresol	0.047	0.034	−0.059	−0.305	0.851	−1.021	0.000	0.000	−0.452	−0.497	−0.045
p-Chlorophenol	0.052	0.043	−0.069	−0.197	0.834	−1.021	0.0000	0.003	−0.354	−0.361	−0.007
Aniline	0.055	0.038	−0.027	−0.492	0.758	−1.021	0.022	0.000	−0.666	−0.675	−0.009
2-Chloroanaline	0.059	0.036	−0.026	−0.393	0.873	−1.021	0.011	0.000	−0.461	−0.458	0.002
p-Nitrophenol	0.062	0.068	−0.085	−0.256	0.882	−1.021	0.000	0.753	0.404	0.401	−0.003
o-Nitrophenol	0.058	0.041	−0.005	−0.364	0.882	−1.021	0.000	0.626	0.218	0.219	0.001
m-Nitrophenol	0.060	0.062	−0.081	−0.226	0.882	−1.021	0.000	0.036	−0.288	−0.241	0.048

[a] $\log k_p$: $\log k$ predicted.
[b] $\log k_e$: $\log k$ experimental.

Table 9
Products of solute descriptors and LSER coefficients for acidic and basic solutes accounting for ionization with the d^+D^+ and d^-D^- terms at 70% methanol.

Solute	*eE*	*sS*	*aA*	*bB*	*vV*	*c*	d^+D^+	d^-D^-	$\log k_p$[a]	$\log k_e$[b]	$\log k_e - \log k_p$
Original											
Pyridine	−0.128	0.614	0.000	−0.518	0.482	−1.281			−0.831	−0.651	0.181
Phenol	−0.163	0.650	−0.091	−0.331	0.553	−1.281			−0.662	−0.755	−0.093
p-Cresol	−0.166	0.636	−0.086	−0.342	0.654	−1.281			−0.585	−0.679	−0.094
p-Chlorophenol	−0.185	0.789	−0.101	−0.221	0.641	−1.281			−0.357	−0.567	−0.210
Aniline	−0.193	0.738	−0.039	−0.552	0.583	−1.281			−0.781	−0.822	−0.041
2-Chloroanaline	−0.209	0.672	−0.038	−0.441	0.671	−1.281			−0.626	−0.651	−0.025
p-Nitrophenol	−0.217	1.256	−0.124	−0.287	0.678	−1.281			0.027	0.571	0.545
o-Nitrophenol	−0.205	0.767	−0.008	−0.408	0.678	−1.281			−0.457	0.330	0.787
m-Nitrophenol	−0.213	1.147	−0.119	−0.253	0.678	−1.281			−0.041	−0.316	−0.275
Modified											
Pyridine	0.026	0.080	0.000	−0.439	0.489	−1.088	0.270	0.000	−0.662	−0.651	0.011
Phenol	0.033	0.085	−0.039	−0.280	0.562	−1.088	0.0000	0.001	−0.726	−0.755	−0.029
p-Cresol	0.034	0.083	−0.037	−0.290	0.664	−1.088	0.0000	0.001	−0.633	−0.679	−0.046
p-Chlorophenol	0.038	0.103	−0.043	−0.187	0.651	−1.088	0.0000	0.006	−0.520	−0.567	−0.047
Aniline	0.039	0.092	−0.017	−0.467	0.592	−1.088	00.0	0.000	−0.779	−0.822	−0.042
2-Chloroanaline	0.043	0.088	−0.016	−0.374	0.681	−1.088	0.005	0.000	−0.661	−0.651	0.011
p-Nitrophenol	0.044	0.164	−0.053	−0.243	0.688	−1.088	0.000	1.066	0.578	0.571	−0.007
o-Nitrophenol	0.042	0.100	−0.003	−0.346	0.688	−1.088	0.000	0.935	0.328	0.330	0.002
m-Nitrophenol	0.043s	0.150	−0.051	−0.215	0.688	−1.088	0.000	0.071	−0.402	−0.316	0.086

[a] $\log k_p$: $\log k$ predicted.
[b] $\log k_e$: $\log k$ experimental.

terms obtained from the unmodified LSER model. It should also be noted that while the positive sign obtained for the s coefficient for the full test set but in the absence of the *D* terms is consistent with other results obtained on this phase, its large value (e.g., 0.49 at 60% and 0.73 at 70% methanol) is not [40,41]. At the mobile phase compositions examined here, the magnitude of the s coefficient on an SCIL phase has been reported to be close to zero [40,41]. This implies that a large part of the ion–dipole type interactions are also incorporated into this term in the absence of the d^+D^+ and d^-D^- terms.

3.4. Evaluation of d^+D^+ and d^-D^- term impact

The d^+ and d^- coefficients are significantly larger than the rest of the coefficients. For this reason, these coefficients may be expected to dominate the retention of the ionizable solutes. However, it is important to remember the contribution to retention for an interaction is the value of the coefficient multiplied by the solute descriptor. The D^+ and D^- solute descriptors are significantly smaller than the solute descriptors for other interactions. To examine the impact of the d^+D^+ and d^-D^- terms on overall retention for the ionizable analytes, Tables 8 and 9 list the products of the solute descriptors and system coefficients for all of the ionizable solutes using the original and modified LSER equations for 60% and 70% methanol, respectively. As can be seen in the tables, only the nitrophenols, aniline and pyridine have any significant *dD* values compared to other LSER terms at 60% and 70% methanol. The remaining ionizable solutes show so little ionization that they could effectively be described by the original LSER model. However, except for *p*-cresol, using 60% methanol, the d^+D^+ and d^-D^- terms of these remaining ionizable compounds still have some small, non-zero contribution to retention. Examination of the differences between the predicted and experimental $\log k$ for these solutes clearly demonstrate that the D^+/D^- modified LSER model does more accurately predict the retention of the more highly ionized solutes than does the original LSER model, as shown in Figs. 7 and 8. Fig. 7 shows the residuals for ionizable solutes when the original LSER model is used while Fig. 8 shows that for the D^+/D^- modified LSER model. The residuals for the ionizable solutes are clearly smaller when the d^+D^+ and d^-D^- terms are included in the model, indicating that the modified model describes their retention better. The residuals for pyridine and 2-chloroaniline overlap in Fig. 8b.

Further evidence of the appropriateness of the modified LSER model may also be ascertained by comparing the experimental and predicted elution order for the ionizable solutes. For instance, without the incorporation of the D^+/D^- modification, the predicted elution order for nitrophenols did not correlate to their actual elution order. The modified model predicts longer retention than with

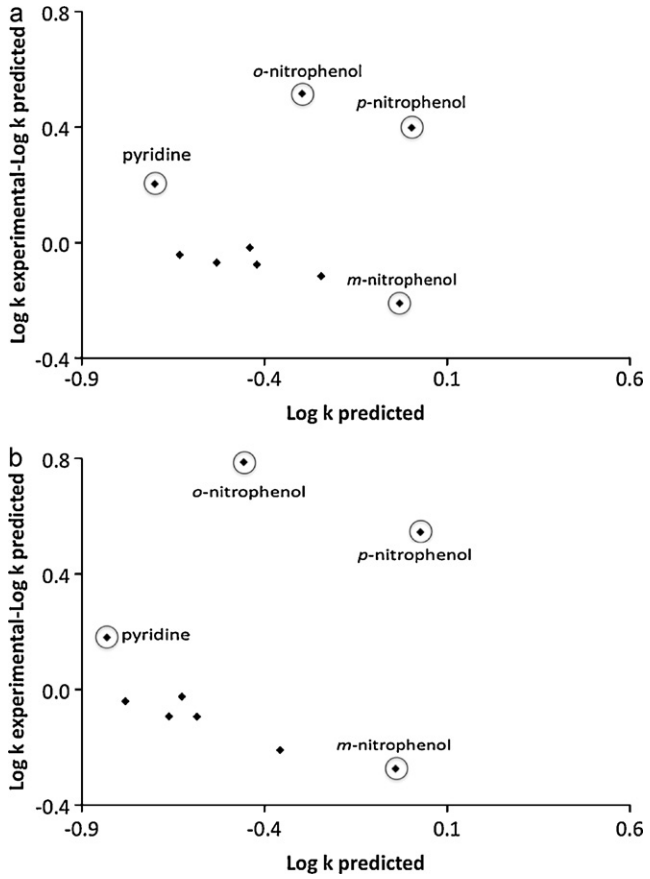

Fig. 7. Plot of residuals ($\log k_{experimental} - \log k_{predicted}$) vs $\log k_{predicted}$ for acidic and basic solutes using the original LSER model at (a) 60% MeOH or (b) 70% MeOH mobile phase composition.

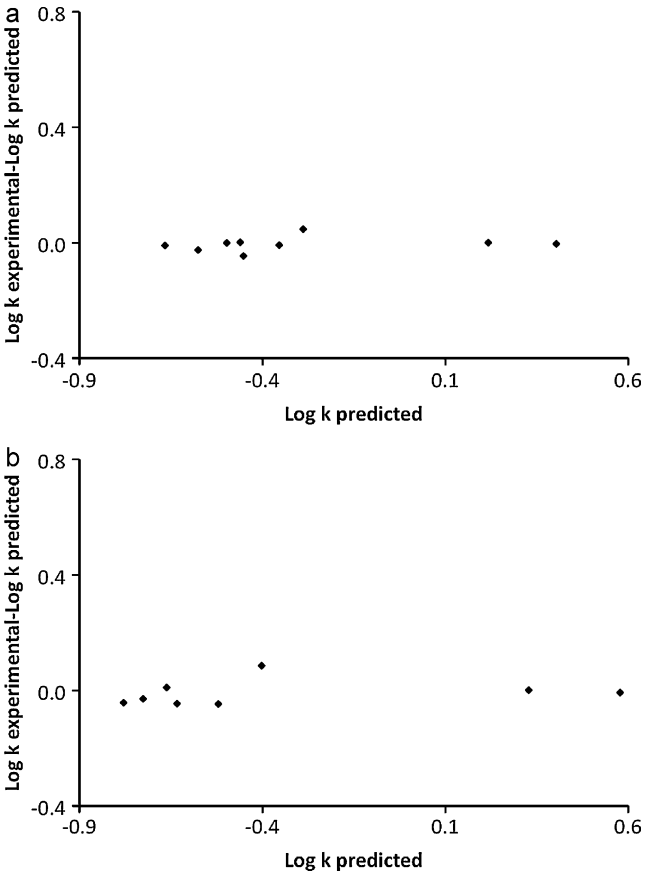

Fig. 8. Plot of residuals ($\log k_{\text{experimental}} - \log k_{\text{predicted}}$) vs $\log k_{\text{predicted}}$ for acidic and basic solutes using the D^+/D^- modified LSER model described by Eq. (5) at (a) 60% MeOH or (b) 70% MeOH mobile phase composition.

the original model when nitrophenols are ionized or partially ionized. In addition, the modified model predicts that the closer their pK_a's to pH of the mobile phase, the stronger attractive interactions between the nitrophenols and the stationary phase. For example, the pK_a's of *o*-nitrophenol, *m*-nitrophenol and *p*-nitrophenol are 8.04, 9.11 and 7.81, respectively. The apparent pH of the 60% methanol/water solution was 7.45. At this pH, it is expected the *p*-nitrophenol would be the most ionized, followed by *o*-nitrophenol, with *m*-nitrophenol being the least. Based on the level of ionization, the elution order should be *m*-nitrophenol, followed by *o*-nitrophenol and *p*-nitrophenol eluting last. The original LSER model predicts an elution order of *o*-nitrophenol, followed by *m*-nitrophenol with *p*-nitrophenol eluting last, which is not correct based on the properties of the nitrophenols and the mobile phase. Hence, the modified model does a better job of predicting the nitrophenol elution order than the original model.

4. Conclusions

The influence of the addition of organic modifiers on the pH of the mobile phase and the ionization of ionizable compounds were considered in characterizing a new ionic liquid n-butylimidazolium bromide based HPLC stationary phase employing a modified linear solvation energy relationship (LSER) approach. Under the mobile phase conditions used in this study, the LSER model modified by the addition of the d^+D^+ and d^-D^- terms was successful in predicting retention characteristics of both neutral and ionizable phenolic compounds on this new phase. The use of the d^+D^+ and d^-D^- terms constitutes an improvement in the fit of the ionizable compounds to the model over that of using a single *dD* term. In addition, the results presented in this work demonstrate that this new stationary phase interacts with solutes simultaneously through both reversed phase and electrostatic interactions.

Acknowledgments

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