Migration behavior of metal complexes in capillary zone electrophoresis
Interpretation in terms of quantitative structure–mobility relationships

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Abstract

Significant advances in metal ion analysis by capillary zone electrophoresis (CZE) have occurred as a consequence of using metal complexes with various organic and inorganic ligands. Metal–ligand complexes that contribute to metal speciation in solution have also gained the attention of researchers. An understanding of the molecular properties that control the separation and further insight into the migration mechanism call for a systematic analysis of relationships between migration parameters and charge and size characteristics of metal complexes. To perform such an investigation, a number multiparametric migration models derived from a generally valid equation for electrophoretic mobility as a function of charge density were developed. The models operating with tabular or readily calculated structural descriptors (in particular, metal atom electronegativity or effective charge) as well as with the formal charge and ligand number were evaluated using numerous sets of experimental migration data for inorganic and organic ligand complexes. Consistent – in a great many instances – approximation results confirm the separation mechanism for metal complexes in CZE as governed basically by differences in charge-to-size parameters, present a valuable and convincing selection of such parameters, possessing a definite physical meaning, and owing to the general validity of the multivariate regression approach, open new possibilities in its application to more complex (i.e., electrokinetic chromatographic) systems. © 2002 Elsevier Science B.V. All rights reserved.

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1. Introduction

The separation and detection of metal ions in the form of precapillary-formed metal complexes has become an accepted practice in capillary zone electrophoresis (CZE) [1–3]. An enhanced separation selectivity, especially compared with that attained for uncomplexed metal ions [4], and substantial improvements in detection, when employing high sensitive derivatization reagents [5] are the major advan-

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tages brought about by this CZE methodology. An overview of the latest advances in CZE as applied to metal ion analysis [6] witnesses that pre-separation derivatization procedures still hold great promise regarding the achievement of multi-elemental separations. Furthermore, distinguishing metal–ligand complexes that govern speciation of a metal in environmental and biological systems seems growing into an important domain of CZE research [7].

Turning to the basic principles of CZE of metal complexes, the state-of-the-art picture does not appear as bright. Whilst a number of modeling approaches and mathematical treatments of systems involving partial on-capillary complex-formation have been reported (see Refs. [4,8,9] for comparison), the migration behavior of metal complexes still lacks quantitative interpretation. Specifically, there are only few attempts made to establish quantitative relationship between migration parameters and intrinsic physical characteristics of metal ions and/or ligand molecular size. Insofar, the most evolved approach relates the electrophoretic mobility of a metal complex to its formal (net) charge, the overall stability constant as a measure for the effective charge and a specific set of ligand structural increments [8,10]. Such an approach, while providing fairly good correlation between experimental and calculated mobilities for certain metal complexes (mostly lanthanide metal chelates of polyaminocarboxylic acids), suffers from uncertainties associated with the stability constants. Also the feasibility of computer-aided modeling based on multivariative regression analysis has not yet been demonstrated for different types of metal complexes. This would ultimately require different sets of informative and quantifiable structural variables in order to provide rigorous mobility approximations.

The present paper intends to examine in detail the migration data for metal complexes available in the literature using the tools of quantitative structure–mobility relationships (QSMRs). The modeling capabilities of derived multiparametric regression equations were assessed with the aim of revealing different predominant properties of metal complexes that determine mobility differences and estimating their importance in CZE. Further extension of iterative regression strategy to the interpretation of migration properties in apparently more challenging electrokinetic chromatographic systems is discussed.

2. General

2.1. Electrophoretic mobility of metal complexes

The electrophoretic mobility of a charged metal complex can be expressed similarly to that of other charged species using a fundamental expression for mobility following from the Stokes–Einstein diffusion model:

$$\mu = q/6\pi\eta r \qquad (1)$$

Eq. (1) relates the mobility of an ionic species (μ) to its charge q, hydrodynamic radius r, and the dynamic viscosity of electrolyte medium η. Strictly speaking, this equation refers to the conditions of infinitive dilution where the effects of ionic strength, acid–base, ion pairing and other secondary interactions are minimal. Also when applying Eq. (1), one should appreciate that no transformation of a given complex in a CZE system (e.g., as a result of dissociation or hydrolysis) takes place or, in case of such transformations, their kinetics is slow enough (compared to the time required for the separation), so that virtually only one metal-containing form exists in each electrophoretic zone (peak).

Rather obvious is the problem of applying Eq. (1) for calculations of the electrophoretic mobility. This problem is due mainly to certain difficulties in estimating the true values of q and r. First, to accurately predict the effect of the metal atom on electrophoretic mobility of the complex, it is necessary not only to know the composition and hence the net charge of the metal complex of interest under CZE conditions but also to account for differences in effective charges for the formally equally charged complexes between different metals and one ligand. However, no data on effective charge calculations for metal complexes in consistent media (like aqueous solutions) could be found in the literature. Therefore, the approximation of q on the basis of a combination of the net charge (Z) and a parameter related to the electron-acceptor ability of the metal atom will be used herein in an attempt to evaluate the migration behavior of similarly charged complexes. These predictors will be discussed in the next section.

In terms of the contribution to the molecular size, the influence of the metal atom on mobility can at a

first approximation be neglected, at least for metal complexes formed by voluminous organic reagents. The size of the ligand plays a substantially greater role and within a series of complexes of a given ligand, its effect can be simply expressed via the ligand number (n_L). The situation becomes more complicated when the estimation of differences in mobility for metal complexes different in the nature of the ligand is the task. According to the principle of molecular dynamics, the size of the ligand (as of any organic molecule) exhibits an additive character and can be presented as a summation of contributions of structural fragments and functional groups. Using the same reasoning, one can try on evaluating the apparent size of both chelate and mixed-ligand inorganic or organic complexes by relying on size-related parameters (see below). Alternatively, the classical theory based on the linearity principle in changes of free energies may be applied to describe how the compositional changes in structurally similar ligands (e.g., upon varying the type of substituent) affect the mobility of complexes.

2.2. Structural descriptors

2.2.1. Parameters of metal atom

Regardless of the type of a complex, a metal atom displays an impact on its effective charge and in the end on the electrophoretic mobility through the mechanism of the electron charge density transfer. The magnitude of such transfer that occurs upon the formation of a metal-electron donor atom bond depends in turn on the electron-acceptor ability of the metal atom. In essence, the greater the effective charge on a metal atom, the higher is the charge on electron donor atoms of a ligand and in case of polydentate (chelating) ligands, on electronegative atoms located away from the coordination polyhedron. For metal complexes formed by acidic chelating reagents considered here as the most representative example, this regularity should lead to the following effect on mobility. Since such chelates bear a negative charge due only to the acid dissociation of one or several functional groups not taking part in the metal ion complexation (the chelate ring is formally electroneutral), they would experience a lesser tend to dissociate (i.e., higher pK_a values) upon increasing the ability of the metal atom to accept electrons. In other words, the metal atom of a

greater effective charge would implicitly provide the anionic chelate complex a lower relative charge and hence a slower mobility. (Note that in this subsection, we assume metal complexes possessing the same net charge as well as the nature and number of ligands).

The electron-acceptor ability can be characterized by a variety of different metal atom parameters (Table 1) from electronegativity χ_M, which is a tabular value, to the effective charge ρ_M, the ratio of electronegativity to ionic radius, orbital electronegativity E_n, the electronic chemical potential μ_M, etc., which are accessible from the specific literature or can be calculated from well-known physical constants (see Section 2.3). All of these were tested and compared as migration models variables, expressing the effective charge of metal complexes, in the course of the present study. Besides, since the assumption of disregarding the metal atom contribution to the molecular size could be incorrect for relatively small inorganic–ligand complexes, the crystallographic radius of the metal ion, r_i was incorporated as a size-related parameter (supplementary to n_L) for modeling their migration behavior.

2.2.2. Ligand parameters

The choice of ligand descriptors used to develop

Table 1
Structural parameters of metal complexes tested for description of the migration behavior in CZE

Parameter	Ref.
Parameters of metal atom	
Electron-acceptor	
Electronegativity	[11]
Effective charge, ρ_M	–[a]
Ratio of electronegativity to ionic radius (χ_M/r_i)	–
Orbital (Klopman) electronegativity, E_n	[12]
Electronic chemical potential, μ_M	[13][a]
Size-related	
Ionic (crystallographic) radius	[14]
Ligand parameters	
Electronic	
Induction constant σ_I	[15]
Resonance constant σ_R	[15]
Size-related	
Number of structural increments, n_i	–
Van der Waals molecular volumes, v	[16]

[a] Calculated as described in Section 2.3.

the migration models required first of all consideration of the expected influence on the apparent size of a metal complex. As mentioned above, the incremental approach has been proven to work fairly well for estimating the relative changes in molecular size with regard to the electrophoretic mobility of certain metal chelates [8,10]. One might have assumed this approach to enable yet better mobility predictions, if the relative size of ligand structural fragments is also taken into account. As the size-determining parameter, Van der Waals molecular volumes of atoms and groups (v) were thought to have been worth assessing. For a few experimental mobility data involving cationic metal chelates of different in the type of substituent but otherwise structurally identical ligands, the electronic constants of substituents were considered as featuring their effect on the distribution of electron density in a molecule.

2.3. Calculations

The effective charges on metal atoms were estimated based on the principle of equalization of electronegativities as a product of the ionic degree of the metal-donor atom (ligand) bond (i) and the metal atom valence number. The i values were calculated from the difference in electronegativity between metal and donor atoms by applying the Pauling equation [17]:

$$i = 1 - e^{-0.18(\chi_M - \chi_L)} \tag{2}$$

where χ_L is the average electronegativity of donor atoms defined as:

$$\chi_L = (\prod_{i=1}^{k} \chi_i)^{1/k} \tag{3}$$

The orbital electronegativities of metal atoms for which data are not available in Ref. [12] were calculated according to Klopman [18] using the tabulated stepwise ionization potentials (I_n and I_{n-1}) and ionic radii obtained from Ref. [13]. The electronic chemical potentials [13] were calculated from the values of the ionization potentials and atomic electron affinities [14].

Calculations of the molecular volume for ionic and molecular ligands (v) were based on the additivity relation [19] which takes into account increments

associated with atoms or groups of atoms (as an average of the data from Refs. [16,20,21]) and the relative stoichiometic multiplicities.

All other parameters are adopted from the literature sources listed in Table 1 or stated below.

2.4. Derivation of migration models, data collection and computation

A basic model expression was obtained by taking the common logarithm of both sides of Eq. (1):

$$\log \mu = a_0 + a_1 \log q - a_2 \log r \tag{4}$$

with both μ and q designated as absolute values. In this equation, the charge density parameters q and r were then expressed in the form of various combinations of selected structural predictors, thus composing a number of multiparametric migration models. The resulting models were applied to fitting of the experimental migration data (electrophoretic or observed mobilities) taken from the literature. Accounting for at minimum a two-parametric character of model equations, data sets comprising ≥ 5 experimental points (i.e., separated complexes) were only accommodated for computation in order to secure the power of performed test.

Migration values were related to separand parameters by means of nonlinear regression processed with the program SigmaPlot 6.0 (Jandel Scientific, Corte Madera, CA, USA) on a Pentium-PC compatible computer. The equations derived were tested according to the requirements of a meaningful correlation analysis by taking into account the coefficient of correlation (R), standard error of the estimate (S.E.), the normality test, the power of a performed test (with the probability of incorrectly concluding that the model is correct, α, set as 0.05), etc., and the number of data points (n) fitted to the equation.

3. Results and discussion

Metal complexes which find application in CZE differ in the charge, ligand type and number, nature of metal–ligand bonding, etc., and depending on the kind of analytes or purpose of the intended separation, may comprise two common combinations: different metals – one ligand and a single metal –

Table 2
Variables of multiparametric regression equations ($\log \mu = a_0 + a_1 \log Z + a_2 \log X_1 + a_3 \log n_L + a_4 \log X_2$)

Metal complexes	Equation	X_1	X_2
Inorganic	5a	χ_M	r_i
	5b	ρ_M	r_i
	5c	χ_M / r_i	–
Organic			
Cationic	6a	χ_M	–
	6b	ρ_M	–
Anionic of 1:1 type[a]	7a	χ_M	–
	7b	ρ_M	–
	7c	χ_M / r_i	–
	7d[b]	E_n	–
	7e	μ	–
Anionic	8a	χ_M	–
	8b	ρ_M	–

[a] $n_L = 1$; that implies a two-parametric form of equations.
[b] Since this parameter exhibits both positive and negative values, the second term of the corresponding regression equation was taken as $a_2 X_1$.

different ligands. Evidently, differences in the composition would make the functional form of Eq. (4) different. For the sake of clearness in model description, metal complexes considered were subdivided into the following classes: (i) inorganic (monode-

ntate) ligand complexes; (ii) cationic chelate complexes; (iii) anionic chelates of a 1:1 type; (iv) anionic complexes of varying ligand numbers; and (v) complexes formed by one metal and variant ligands (but exhibiting no heterogeneity with respect to the type of electron-donor atoms as those are not yet available within a single CE experimental series). Variables of the migration models subjected to evaluation in this project are collected in Table 2. It should be noted at this point that variables entry (in the form of combinations Z–X_1 and n_L–X_2) and functional dependence of model equations are not quantitatively accounted for by any theoretical relation but empirically defined.

3.1. Inorganic complexes

Separations of complexes of inorganic ligands that include a representative number of metal ions encompass chloro [22–24], bromo [25] and cyano [26–28] complexes. Table 3 shows the performance of migration models operated with Eqs. (5a)–(5c) which were verified with the respective data sets. As might be expected, the four-parametric equations, i.e., Eqs. (5a) and (5b), cannot be used to adequately represent the experimental results containing smaller

Table 3
Statistical criteria for nonlinear regression of model Eqs. (5a)–(5c) to experimental data for metal–inorganic ligand complexes

Ligand	Metal ions	n	Equation	Regression coefficients				R/S.E.	Ref.
				Log Z	Log X_1	Log n_L	Log r_i		
Chloride	Ir, Os, Pd, Rh, Ru	5	5c	1.000	4.708	−1.395	–	0.9993 0.011	[22]
	Ir, Os, Pd, Pt(II), Pt(IV), Rh	6	5a	1.206	1.805	−0.026	−6.495	0.9894 0.040	[23]
	Au, Ir, Os, Pd, Pt, Rh	6	5a	0.716	3.932	−0.321	−2.664	0.9670 0.061	[23]
	Platinum metals and Au	19[a]	5b	0.811	1.299	−1.544	−1.999	0.9401 0.056	[24]
Bromide	Au, Ir, Pd, Pt, Rh	5	5c	1.342	3.102	−1.629	–	–[b]	[25]
Cyanide	Cu, Fe, Ni, Pd, Pt	5	5c	0.600	0.560	−0.652	–	0.9861 0.040	[26]
	Co, Cr, Fe(II), Fe(III), Pd, Pt	6	5a	1.251	2.313	−1.129	−0.662	0.9499 0.060	[27]
	Co, Cr, Cu, Fe(II), Fe(III), Ni, Pd, Pt	8	5a	1.008	1.418	−0.689	−0.034	0.9880 0.021	[28]

[a] Including mixed-ligand aqua and hydroxyaqua complexes.
[b] The power of the performed test is below the desired power.

numbers of points. Therefore, these data sets were reapplied to Eq. (5c) accounting the metal atom size indirectly, as such included in χ_M/r_i. However, there was seldom a considerable improvement in the correlation. For other data presented, agreement between the experimental and calculated mobilities appeared satisfactory ($R > 0.96$), except for platinum metal mixed-ligand complexes [24] and metal cyanides separated under conditions where distinct ion-pairing interactions in a CZE system take place [27] ($R > 0.94$). In the former case, this may be attributed to the fact that the present model is not capable to differentiate between Cl^-, H_2O and OH^- ligands regarding their contribution in the molecular size. For this reason, the n_L value for the complexes of Ru and Rh, existing in the carrier electrolytes employed as aquapentahalogenides, was assigned to six.

Appealing to the coefficients relating log μ and specific adjustable parameters, it should be noted that while the signs at log Z (positive) and log n_L and log r_i (both negative) follow logically from Eq. (1), relationship between the mobility and electron-acceptor parameters of the metal atom (X_1 in Table 2) requires comments. The positive influence of both χ_M and χ_M/r_i indicates that complexes with the electron charge density shifted more strongly to the central atom have slower mobilities. As this might only be assigned to a decrease in the effective (negative) charge of separands under consideration, more positively charged metal atoms appear favoring such a decrease, the regularity being not trivial for an a priori prediction. Here one must note again that measuring the effective charges in metal complexes, even in solid state, remains a challenging task. Yet available the results may exhibit an unlooked-for character. In metal cyanides, for instance, the metal atom was found to possess a negative charge stemming from substantially covalent metal–cyano bonds [29]. This makes speculations about the metal atom effect on the mobility often complicated.

Also worthwhile mentioning is the radius of the metal ion as a more marked factor controlling the electrophoretic mobility of chloride complexes relative to the size of the ligand part of a complex (in terms of average values of the corresponding coefficients; a_3 and a_4 are -0.63 and -3.72, respec-

tively). This observation has an opposite trend for metal–cyano complexes that is in accord with a greater relative size (volume increment) of the cyanide ion (31.3 and 28.9 Å^3 [19], respectively) and conforms our earlier discussion (see Section 2.2).

3.2. Cationic chelates

Metal complexes of this type are limited to transition metal chelates formed by bidentate neutral ligands like 2,2′-bipyridine (bpy) [30,31], 1,10-phenanthroline (phen) [31] or their derivatives [31,32]. Also exhibiting a positive net charge are complexes of certain acidic reagents (e.g., substituted azobenzenes containing no ionizable functional groups [33,34]) with the number of (monobasic) chelating ligands smaller than the metal ion charge. Migration models describing log μ of these complexes were reduced to three-parametric equations (Eqs. (6a)–(6c) in Table 2) by omitting the metal ion radius term on the grounds of dominating contribution of a ligand(s) in the molecular size. Eqs. (6a)–(6c) fitted to the experimental data offered, in the majority of calculations, good agreement with theoretical results ($R > 0.98$); a somewhat better quality of fit was provided by Eq. (6a) (i.e., operated with χ_M). As was the case for inorganic–ligand complexes, the positive sign at χ_M (and correspondingly the negative influence of ρ_M) can unambiguously be interpreted as a faster mobility of complexes originated from the metal atom having a lower ability to accept electrons. The only reason for such migration behavior which may be advanced here is that the metal atom seems to be the only center of the localized positive charge in the molecule; changes in this charge only would lead to changes in mobility observed.

As a certain practical limitation of QSMRs manifested, apprehension of the actual composition of metal complexes in an electrolyte environment should be emphasized. For example, coordination of the acetate ions to $[Cu(bpy)_2]^{2+}$ and $[Cu(phen)_2]^{2+}$ was proven to occur in acetate buffer electrolytes [32]. This explains why satisfactory predictability of model equations was only attained when copper complexes were taken in computation as a single-charged species.

3.3. Anionic chelates of a 1:1 type (aminpolycarboxylates)

Aminopolycarboxylic acids comprise the group of chelators most frequently used in CZE analysis. The abundance of literature data including typically a broader range of metal ions separated allowed a wider set of metal atom parameters as the mobility predictors to be explored (see Table 2). Among these, χ_M/r_i assessed with three representative data sets ($n = 9$–14) for EDTA complexes showed a poorer correlation with log μ than χ_M and ρ_M did, and for this reason was not considered further. The application of the electronic chemical potential is restricted by the fact that the values μ_M are available for integral values of the metal oxidation state, which actually do not occur in the complexes. The orbital electronegativity (metal softness parameter) describes the degree of the electron density transfer from electron donor atoms to metal cations of soft and intermediate (by Pearson) type. This is the rationale

why the statistically significant correlations between log μ and E_n were only found with migration data sets containing no alkaline earth metal or aluminum (III) complexes. The best-fit equation:

$$\log \mu_{eff} = 1.355 + 0.922\,(\pm 0.020)\log Z$$
$$+ 0.013\,(\pm 0.002)E_n$$
$$R = 0.9986;\ \text{S.E.} = 0.009;\ n = 9 \tag{9}$$

(the values in parentheses are standard deviations of regression coefficients) gave for a range of metal–CDTA complexes [35] an approximation comparable in goodness to those obtained by means of Eq. (7a) or (7b) (see Table 4). Nonetheless, only parameters χ_M and ρ_M were selected for the subsequent calculations as those characterized by higher correlation factors irrespective of the nature of the metal atom.

The results from a two-parametric nonlinear regression analysis undertaken using Eqs. (7a) and (7b) are shown in Table 4. For all of the fits obtained (with only exception) the regression coefficient at the

Table 4
Selection of the most meaningful models for metal–aminopolycarboxylic acid complexes

Ligand[a]	Metal ions	n	Equation	Regression coefficients		R/S.E.	Ref.
				Log Z	Log X_1		
EDTA	Alkaline earths, transition metals, Pb	14	7a	$-$[b]	0.329	0.9622 0.012	[36]
	Cd, Co(II), Cr(III), Fe(III), Ni	5	7a	0.107	0.854	0.9744 0.010	[37]
	Co(II), Cu, Fe(III), Ni, V(IV), V(V)	6	7a	0.701	0.499	0.9879 0.022	[38]
	Ba, Ca, Cu, Fe(III), Mn, Zn	6	7a	0.543	0.177	0.9960 0.007	[39]
CDTA	Alkaline earths, transition metals	8	7b	0.524	-0.221	0.9970 0.005	[40]
	Transition metals, Bi, Pb	9	7b	0.901	-0.214	0.9910 0.022	[35]
	Alkaline earths, transition metals, Pb	10	7a	0.438	0.100	0.9757 0.014	[41]
DTPA	Transition metals, Bi, Pb	8	7b	0.689	-0.257	0.9767 0.005	[35]
HBED	Alkaline earths, transition metals, Al	12	7b	0.686	-0.040	0.9883 0.017	[42]

[a] CDTA = *trans*-1,2-cyclohexanediaminetetraacetic acid; DTPA = diethylenetriaminepentaacetic acid; HBED = *N*,*N*′-di(2-hydroxy-benzyl)ethylenediamine-*N*,*N*′-diacetic acid.
[b] Z is constant for M(II) complexes under examination.

term log Z is substantially larger in magnitude than that at the logarithm of either metal atom parameter (on average 0.640 and 0.215, respectively). Since the individual scale of both parameters is similar, this allows for a conclusion that the net charge of the metal complex has a greater impact on mobility. Among the complexes sharing a common net charge, those formed by a more electronegative metal atom, as predicted, have a higher effective charge, causing them to migrate more quickly (unless the counter-migration system is utilized). Accordingly, a negative effect of ρ_M was encountered in all the cases of statistically relevant correlations.

Fig. 1 is a correlation plot for the nine metal–CDTA complexes ($R > 0.99$). A closer examination of the data revealed two clusters of points belong to M(II) and M(III) complexes fairly different in mobility. This observation leads one to conclusion (as correctly noted both anonymous reviewers) that within the group of different M(II) or M(III) complexes the model equation predicts the observed mobility less accurately. Nonetheless, in general fitted log μ values agree quite well with experimental data: the average difference between observed
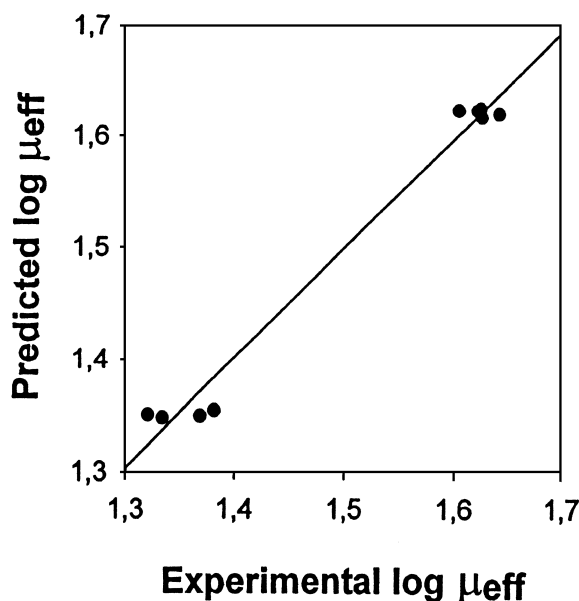

Fig. 1. Relationship between the migration parameters of metal–CDTA complexes determined experimentally [35] and calculated using Eq. (7a). Slope 0.981, intercept 0.029, R 0.9904, the average deviation of the calculated and observed log μ_{eff} values is 1.07%.

and calculated values log μ is 0.016 log units with the largest residual of 0.032. It is interesting to note that the value of χ_M for V(IV) required for this and several other calculations is missing in the literature. Of two possible options, using an average of the corresponding values for V(III) and V(V) or evaluating the inaccessible parameter from CZE data, the latter resulted in better fits.

3.4. Anionic chelates

Metallochromic and similar organic reagents form an important group of complexing ligands which have been increasingly applied to separations of metal ions due to the advantage of offering very sensitive photometric detection [1,5]. Table 5 lists a representative number of these ligands suitable for computation as having effected the sufficient number of metals separated. It should be noted, however, that extra functionalities in metallochromic ligands essential for providing the metal complexes an enhanced absorptivity cause the separands to become larger and differences in the electrophoretic mobility less significant. Therefore, the manipulation of separation selectivity has to be devised [1,4], and approaches involved can include ion-pair interactions [45,49], an auxiliary complexation with a competing ligand [47], introducing a micellar phase [50], use of on-capillary complexation mode [48], etc. (all the examples herewith are given for the systems subjected to regression). This makes questionable whether the basic Eq. (1) remains valid under such circumstances (see Section 2.1). From the data of Table 5, it can be seen that the modeling approach employed survives – with minor exceptions [45,49,50] – secondary interactions in a CZE system. The exceptions mentioned concern in particular the effect of certain ion pairing additives on the migration behavior of metal complexes of 4-(2-pyridylazo)resorcinol (PAR) [49] and 2,2′-dihydroxyazobenzene-5,5′-disulfonic acid (DHABS) [45]. As a result, a worse agreement between the experimental and predicted mobilities ($R < 0.95$) was obtained.

Apart from the complexes listed, several other complexing systems were tested with the result of statistically less significant models based on using Eqs. (8a) and (8b). Whilst it may be speculated what is the reason of less consistent correlations for rather

Table 5
Summary of multiple regression analysis (Eqs. (8a)–(8c)) for anionic metal–chelate complexes

Ligand[a]	Metal ions	n	Equation	Regression coefficients			R/S.E.	Ref.
				Log Z	Log X_1	Log n_L		
PAR	Co(III), Cu(II)[b], Fe(II), Ni(II), V(V)	5	8b	0.510	−0.345	−0.115	0.9707 0.023	[43]
	Cd(II), Co(III), Cu(II)[b], Fe(II), Ni(II), Zn(II)	6	8b	0.553	−0.116	−[c]	0.9976 0.006	[44]
DHABS	Al(III), Co(III), Cr(III), Cu(II), Fe(III), V(V)	6	8a	1.450	0.219	−1.318	0.9974 0.010	[45]
BPS	Co(II), Cu(II), Fe(II), Ni(II), Zn(II)	5	8b	0.302	−0.112	−0.114	0.9895 0.008	[46]
Arsenazo III	Ce(III), La(III), lanthanides, U(VI)	15	8c[d]	1.338	1.052	−[c]	0.9789 0.007	[47]
PDC	Co(II), Cu(II), Fe(II), Fe(III), Ni(II)	5	8a	0.207	0.411	−[c]	0.9737 0.009	[48]

[a] BBS = bathophenanthrolinedisulfonic acid; DHABS = 2,2′-dihydroxyazobenzene-5,5′-disulfonic acid; PAR = 4-(2-pyridylazo)resorcinol; PDC = 2,6-pyridinedicarboxylic acid.

[b] The copper–PAR complex tends to vary the composition from 1:1 to 1:2 over the pH of the electrolyte buffer utilized. Both options were taken into regression and according to the quality of fit, the chelate with $Z = 2$ (i.e., $[Cu(PAR)_2]^{2-}$) is predominant in the CZE process.

[c] The same metal-to-ligand ratio for all complexes.

[d] Log $\mu = a_0 + a_1 \log Z - a_2 \log (\chi_M/r_i) - a_3 \log n_L$.

simple complexes of 8-hydroxyquinoline-5-sulfonic acid [51,52] and 3-nitroso-1-naphthol-4-sulfonic acid [53], a poor regression derived for metal chelates of Alizarin complexone [54] was not unexpected. A negative charge of these complexes is due to the dissociation of H_2O coordinated with the metal atom that could give rise to its unusual effect on mobility (compared to that described in Section 2.2).

3.5. Complexes of a single metal and various ligands

A number of CZE studies have been focused on the separation of different ligand complexes of the same metal atom. Differences in the mobility of such complexes should be ascribed to differential charge densities arising from differences in the structure of organic part of the molecule. As pointed out in Section 2, variations in the nature and the number of ligands or their functional groups impart both different charges and sizes to the metal complex. In an attempt to establish QSMRs, two basic migration models represented by the following expressions:

$$\log \mu = a_0 + a_1 \log Z - \sum (a_i n_i) \tag{10}$$

$$\log \mu = a_0 + a_1 \log Z - a_2 \sum (n_i v_i) \tag{11}$$

were applied to available mobility data. As can be seen, Eq. (10) takes into account the increments of the various functional groups (atoms) into the molecular size (each with its own regression coefficient), whereas the relative size of the ligand structural fragments is additionally included in the integral size-related term of Eq. (11).

For a series of inorganic cobalt(III) coordination compounds (mostly cobaltammines) [55], running a nonlinear regression analysis with tabular or calculated v values for molecular (ammonia, water, ethylenediamine) and ionic (chloride, carbonate, oxalate) ligands resulted in a fairly good agreement between experimentally measured and calculated log μ ($R > 0.98$). On the other hand, none of fits performed using the two sets of copper [56] and vanadium [38] complexes with different aminopolycarboxylic ligands permitted statistically satisfactory regressions, no matter what the type of equation was fitted. This

observation is deemed surprising as the influence of the size of these ligands on mobility was earlier proven to be well quantified in terms of increments of carboxylic, methylene and tertiary amino groups [8,10]. A further data set comprising many metal complexes of EDTA, *trans*-1,2-cyclohexanedi-aminetetraacetic acid (CDTA) and diethylenet-riaminepentaacetic acid (DTPA) [35] was therefore used for determining the validity of a three-parametric migration model:

$$\log \mu = a_0 + a_1 \log Z + a_2 \log \chi_M - a_3 \sum(n_i v_i)$$
(12)

including the same size-dependent term. This combined equation describes the linear change in log μ as a function of charge and ligand structure characteristics taken in computation simultaneously. Fig. 2 shows the result of applying Eq. (12) to all 24 complexes. The linear relationship obtained emphasizes the appropriateness of molecular volumes for rather precise predictions of log μ in situations where variant ligand complexes are encountered.
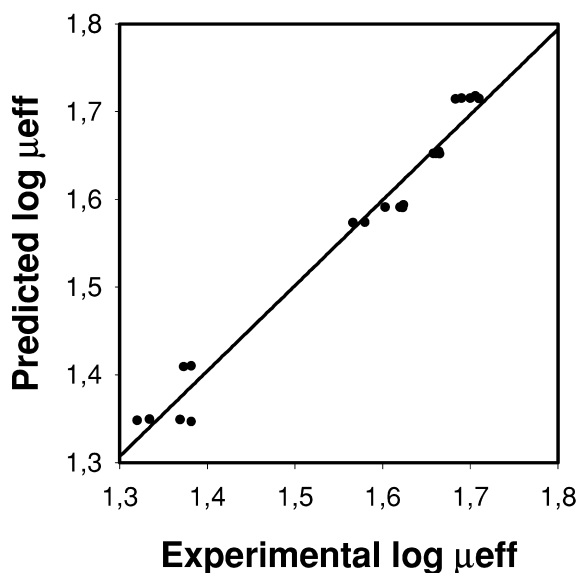

Fig. 2. Correlation plot for aminopolycarboxylic complexes of Bi(III), Co(II), Co(III), Cr(III), Cu(II), Fe(III), Hg(II), Ni(II) and Pb(II). Slope 0.974, intercept 0.041, correlation coefficient, 0.9869. Predicted values were obtained from equation: $\log \mu_{eff} = 2.770 + 0.808 \, (\pm 0.032) \log Z + 0.053 (\pm 0.280) \log \chi_M - 0.575 (\pm 0.103) \log \sum(n_i v_i)$ (S.E. = 0.025; $n = 21$).

The last migration data underwent modeling approximation in this work involved the two sets of μ values for cationic copper (II) chelates with substituted 1,10-phenanthrolines [31,32]. The effective positive charge of these complexes is localized on the metal atom (see Section 3.2), and hence the mobility was thought to be dependent on the electronic effects of substituents (as well as on their size) in the following manner:

$$\log \mu = a_0 - a_1 \sum \sigma_I - a_2 \sum \sigma_R - a_3 \sum(n_i v_i) \quad (13)$$

where σ_I and σ_R are induction and resonance constants, respectively, and a_0 incorporates also the net charge Z which is constant for all of the complexes under examination. In this equation, a_2 and a_3 stand having negative signs as the shift of the electron density from the ligand toward the metal atom due to either effect should translate into a lower mobility of the complex.

The use of Eq. (13) was however circumscribed by the fact that the corresponding electronic constants are scarcely available for substituents in heterocyclic conjugated molecules. Estimations within the formalism of the theory of unbinding molecular orbitals [57] appear to be a nontrivial task and were not attempted here. Alternatively to cope with this complication, we applied the classical Hammett equation expressing the sum of substituent constants as a difference between the logarithms of equilibrium constants for substituted and non-substituted compounds, i.e., $\log K - \log K_0$. Using this expression, the right-hand side of Eq. (13) will read:

$$\log \mu = a_0 + a_1 \cdot (\log K - \log K_0) - a_2 \sum(n_i v_i)$$
(14)

Taking the Cu(II) complex stability constants or acid dissociation constants of the phenanthrolines as K (K_0 is the respective constant for $[Cu(phen)_2]^{2+}$ and phen) [58], the experimental data were fitted to Eq. (14). Still the power of performed regressions was above the desired power for only one of four fits. The coefficient of correlation of this fit was 0.9004 [$\log \mu_{obs} = 1.852 + 0.0278 \, (\pm 0.0080) \, (\log K_a - \log K_{a(0)}) - 0.0009 \, (\pm 0.0002) \sum(n_i v_i)$; S.E. = 0.017; $n = 7$]. A possible explanation for this incomplete agreement may be a different way how changes

in the distribution of electron density in a ligand exert an influence on complex formation or acid dissociation on the one hand and on electrophoretic movement of the resultant complex on the other.

4. Conclusions

The multivariate regression approach for analyzing structure–mobility dependencies adequately described and rationally interpreted CZE data for various metal complexes, and this is the key conclusion drawn from this study. Operating with a number of meaningful migration models, this strategy also contributes to a better understanding of migration behavior of metal complexes. For instance, some variations in the adjustable parameters indicate that along with the sole electrophoretic effect, there occur secondary interactions influencing migration of metal complexes in CZE. As a matter of fact, these interactions should cause the compositional changes of migrating species. Provided that the high statistical quality of the model approximation was achieved, it is possible to solve the reverse problem – that is, to determine actual structural parameters and then the composition of the metal complex from the back-calculations of CZE data.

Harmonious approximations of migration achieved from regression equations derived here give rise to expectation that QSMRs applicable to more complex CE modes, e.g., electrokinetic chromatographic systems, could as well be advanced. The separation mechanism in such systems is governed chiefly by electrophoretic migration and hydrophobic or ion-exchange interactions. The success in developing the corresponding migration models will therefore depend on our ability to identify the analyte parameters that significantly influence the migration due to both contributions and could be then taken into computation with the result of accurate predictions. This is the aim of our forthcoming investigations.

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