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Medium Effect on the Acid−Base Properties of Branched Polyethylenimine in Different Aqueous Electrolyte Solutions†

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The effect of different ionic media on the protonation of branched polyethylenimine (BPEI) was studied potentiometrically in aqueous solutions containing LiCl, KCl, and Me_4NCl from (0.5 to 3) $mol\cdot L^{-1}$ and $CaCl_2$ from (0.2 to 2) $mol\cdot L^{-1}$ at $T = 298.15$ K. Protonation constants of BPEI in alkali metal chlorides are quite similar, and the trend is (alkali Cl) > $CaCl_2$ > Me_4NCl. The dependence on ionic strength of K^H, in different ionic media, was modeled using the Specific ion Interaction Theory (SIT) approach. Differences in protonation constants (in $CaCl_2$ and Me_4NCl, at low ionic strength, $I < 1\ mol\cdot L^{-1}$) were also interpreted in terms of weak complex formation. These complexes are quite weak: log K (M + A = MA) = (24.9 and 24) $L\cdot mol^{-1}$, for M = Ca^{2+} and Me_4N^+ (A = BPEI), respectively, at $I = 1\ mol\cdot kg^{-1}$. BPEI protonation enthalpies and entropies were determined by direct calorimetric titrations at different ionic strengths. The ΔH_i trend is KCl > NaCl > LiCl > $CaCl_2$, for each protonation step of BPEI. Dependence on ionic strength for ΔH_i was taken into account by the SIT model.

Introduction

Branched polyethylenimine (BPEI) is a water-soluble polymer. Its polymeric structure presents a high concentration of polar groups, with a nominal ratio of primary to secondary to tertiary amines reported as 0.25:0.50:0.25. Its monomeric unit is shown in Figure 1. These four different nitrogen atoms, present in each monomeric unit, are protonable at different pH, and for this reason, the BPEI is considered as a "proton sponge" virtually at any pH value. BPEI is employed in many fields for its binding properties toward metal ions[1−6] and organic compounds (ref 7 and references therein). It is widely used in industrial fields for wastewater purification (ref 8 and references therein), for example, in paper industries,[9−11] and fractionation of hydrometallurgical process streams (ref 12 and references therein). Moreover, BPEI is employed to form an ionically conducting system with metal salts utilized as electrolytes (ref 13 and references therein) and to prepare mono- or multilayer films[14−16] with various *n*-alkanoic acids. For its biocompatibility and transfection efficiency, BPEI and its derivatives have a significant role in DNA condensation as synthetic gene carriers in nonviral gene delivery systems.[17−22] This is due to its capacity to condense DNA in small and reproducible particles that can be transported in the cell escaping the lysosomal compartment.[23−25] This peculiarity makes BPEI clinically helpful for gene therapy and diseases such as cystic fibrosis[26−29] and lung cancer.[26,30,31]

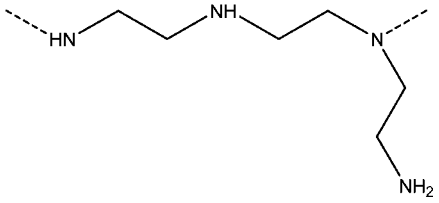

Figure 1. Representation of the BPEI monomeric unit.

Table 1. Experimental Details of Potentiometric Measurements, at $T = 298.15$ K

	I / $mol\cdot L^{-1}$	C_{BPEI} / $mmol\cdot L^{-1}$	pH
Li	0.53 to 2.82	4 to 9	2.2 to 11.0
K^+	0.52 to 2.83	5 to 10	2.1 to 10.9
Ca^{2+}	0.51 to 5.58	4 to 9	1.8 to 10.3
Me_4N^+	0.51 to 2.82	4 to 10	1.9 to 10.4

Although BPEI is so important in many scientific and technical fields, only few papers regarding its acid−base proprieties[32−34] are present in the literature, and many of these report no thermodynamic parameters or quantitative data. In a previous work,[35] we studied the protonation equilibria of BPEI in NaCl aqueous solutions, and we proposed a valid approach to process the pH potentiometric data. This method considers the BPEI as a molecule having four independent protonable groups, and only the third protonation step is analyzed as a function of the protonation degree, $\bar{p}$. In the present work, we checked this method, by analyzing the potentiometric data obtained in four different ionic media (LiCl, KCl, $CaCl_2$, and Me_4NCl) at different ionic strengths as reported in Table 1. During this investigation, we observed that protonation constants of BPEI have a trend similar to some low molecular weight amines already studied[36−38]

$$\text{alkali metal chlorides} > CaCl_2 > Me_4NCl$$

and this trend is quite different compared with carboxylic or inorganic anions,[39,40] for which the trend is alkali metal ions $\gg Ca^{2+}$ and $Me_4N^+ \gg$ alkali metal ions. This trend is due to the formation of weak species between BPEI and Ca^{2+} and Me_4N^+ cations of the supporting electrolyte. Therefore, we have

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Table 2. Intrinsic Protonation Constants of BPEI in Different Supporting Electrolytes, at $I \approx 0.5$ (mol·L^{-1}) and $T = 298.15$ K, According to Equation 4

I	$\log K^{H}_{n1}$	$\log K^{H}_{n2}$			$\log K^{H}_{n4}$	
mol·L^{-1}	$n_1 = 1$	$n_2 = 1$	$\log K^{H}_{n3}$	n_3	$n_4 = 1$	σ^a
			LiCl			
0.529	9.902 ± 0.006[b]	8.650 ± 0.015[b]	6.224 ± 0.014[b]	1.696 ± 0.040[b]	2.538 ± 0.014[b]	0.0517
			KCl			
0.515	9.802 ± 0.017	8.579 ± 0.003	6.221 ± 0.008	1.519 ± 0.021	2.524 ± 0.006	0.0270
			$CaCl_2$			
0.513	9.582 ± 0.008	8.317 ± 0.011	5.954 ± 0.009	1.534 ± 0.033	2.479 ± 0.013	0.054
			Me_4NCl			
0.514	9.735 ± 0.008	8.502 ± 0.007	6.131 ± 0.006	1.517 ± 0.013	2.497 ± 0.008	0.0287

[a] Standard deviation on the fit. [b] ± Standard deviation.

Table 3. Protonation Constants of BPEI in Different Supporting Electrolytes, at $I \approx 0.5$ mol·L^{-1}, Using Different Models, at $T = 298.15$ K

I / mol·L^{-1}		$\log K^{H}_{1}$	$\log K^{H}_{2}$	$\log K^{H}_{3(1)}$[a]	$\log K^{H}_{3(0)}$[a]	$\log K^{H}_{3(m)}$[a]	$\log \bar{K}^{H}_{3}$	$\log K^{H}_{4}$	σ^b	σ^2/σ_0^{2c}
						LiCl				
0.529	m1	9.789 ± 0.008[d]	8.758 ± 0.006[d]				6.252 ± 0.026[d,e]	2.530 ± 0.016[d]	0.056	1.306
	m2	9.802 ± 0.009	8.693 ± 0.014	6.796 ± 0.037[d]	5.694 ± 0.025[d]		6.245	2.5354 ± 0.015	0.049	1.000
	m3	9.767 ± 0.075	8.646 ± 0.028	6.869 ± 0.053	5.740 ± 0.068	6.173 ± 0.058[d]	6.261	2.5331 ± 0.015	0.049	1.000
						KCl				
0.515	m1	9.692 ± 0.009	8.688 ± 0.005				6.258 ± 0.017	2.496 ± 0.009	0.0476	1.586
	m2	9.705 ± 0.007	8.634 ± 0.010	6.711 ± 0.026	5.781 ± 0.030		6.246	2.498 ± 0.008	0.0388	1.054
	m3	9.700 ± 0.007	8.665 ± 0.012	6.628 ± 0.046	5.640 ± 0.080	6.378 ± 0.064	6.215	2.504 ± 0.009	0.0378	1.000
						$CaCl_2$				
0.513	m1	9.485 ± 0.010	8.396 ± 0.021				6.005 ± 0.019	2.455 ± 0.005	0.050	1.487
	m2	9.496 ± 0.008	8.342 ± 0.011	6.428 ± 0.025	5.521 ± 0.027		5.975	2.462 ± 0.008	0.042	1.049
	m3	9.494 ± 0.009	8.363 ± 0.009	6.371 ± 0.072	5.438 ± 0.088	6.046 ± 0.081	5.951	2.466 ± 0.009	0.041	1.000
						Me_4NCl				
0.514	m1	9.639 ± 0.002	8.619 ± 0.007				6.193 ± 0.014	2.477 ± 0.006	0.040	1.563
	m2	9.651 ± 0.005	8.565 ± 0.012	6.634 ± 0.039	5.698 ± 0.039		6.166	2.480 ± 0.005	0.033	1.063
	m3	9.644 ± 0.003	8.605 ± 0.010	6.500 ± 0.058	5.528 ± 0.079	6.356 ± 0.071	6.128	2.490 ± 0.009	0.032	1.000

[a] Values calculated from the Högfeldt three-parameter equation. [b] Standard deviation on the fit. [c] Variance ratio. [d] ± standard deviation. [e] $\log K^{H}_{3}$ values obtained from eq 7.

described this interaction by both using the SIT approach[41−43] and introducing a model that takes into account the formation of weak species. The literature does not report any work about the interaction of BPEI toward Ca^{2+} or Me_4N^+. Recently, the interaction between BPEI and the phytate anion was quantitatively studied.[44]

As a further contribution to BPEI acid−base properties in different aqueous media, the results of a calorimetric investigation on BPEI protonation in solutions containing the most important inorganic salts in natural water and biological fluids (LiCl, NaCl, KCl, and $CaCl_2$) are reported in this paper. The direct calorimetric titrations were performed at 298.15 K, at different ionic strengths for each ionic media. The SIT equation was applied to modeling the dependence of protonation enthalpies on ionic strength for each ionic media.

Experimental Section

Chemicals. Hydrochloric acid and sodium hydroxide solutions were prepared by diluting concentrated ampoules (Riedel-deHaën) and were standardized against sodium carbonate and potassium hydrogen phthalate, respectively. LiCl and KCl aqueous solutions were prepared by weighing pure salts (Fluka) dried in an oven at 110 °C. Tetramethylammonium chloride [Me_4NCl] aqueous solution was prepared by weighing pure salt (Fluka) previously purified by recrystallization from methanol and vaccuum-dried. Calcium chloride solutions were prepared from the *puriss.* solid product (Fluka), previously vacuum-dried, and then standardized by EDTA complexometric titrations. BPEI solutions were prepared by diluting a 50 % aqueous solution of BPEI MW 750 kDa (Aldrich) without further purification. Their concentrations were checked potentiometrically by alkalimetric titrations. All solutions were prepared with analytical grade water ($R = 18$ MΩ·cm^{-1}) using grade A glassware.

Apparatus. Potentiometric titrations were carried out at (298.15 ± 0.01) K using an apparatus consisting of a model 713 Metrohm coupled with a combination ISE-H Orion Ross type glass electrode model 8172 and a model 765 Metrohm motorized burette. The estimated precision was ± 0.15 mV and ± 0.003 mL for the voltage and titrant volume readings, respectively. The apparatus was connected to a PC, and automatic titrations were performed using a suitable computer program to control titrant delivery and data acquisition and to check for emf stability. To exclude O_2 and CO_2 inside, all titrations were carried out under magnetic stirring, and bubbling purified presaturated N_2 through the solution. Calorimetric measurements were carried out at (298.150 ± 0.001) K using an apparatus consisting of a model 450 Tronac isoperibol titration calorimeter coupled with a Keithley 196 system digital multimeter. Titrant was delivered by a 2.5 mL capacity Hamilton syringe (model 1002TLL). The apparatus was connected to a PC, and automatic titrations were performed using a suitable computer program to control calorimetric data acquisition. The uncertainty of the calorimetric apparatus was checked by titrating THAM [tris(hy-

Table 4. Protonation Constants of BPEI in Different Supporting Electrolytes, at Different Ionic Strengths, in the Molal Concentration Scale, at T = 298.15 K

$\frac{I}{\text{mol}\cdot\text{kg}^{-1}}$	log K^{H}_{1}	log K^{H}_{2}	log $K^{H}_{3(1)}$ [a]	log $K^{H}_{3(0)}$ [a]	log $\bar{K}^{H}_{3}$ [b]	log K^{H}_{4}
			LiCl			
0.54	9.796 ± 0.009[c]	8.687 ± 0.014[c]	6.790 ± 0.037[c]	5.688 ± 0.025[c]	6.239	2.529 ± 0.015[c]
1.05	9.918 ± 0.010	8.941 ± 0.018	7.024 ± 0.057	6.005 ± 0.048	6.514	2.602 ± 0.012
1.97	10.102 ± 0.008	9.187 ± 0.020	7.376 ± 0.046	6.149 ± 0.057	6.763	2.731 ± 0.015
2.99	10.302 ± 0.012	9.401 ± 0.017	7.649 ± 0.036	6.227 ± 0.039	6.938	2.825 ± 0.021
			KCl			
0.52	9.698 ± 0.007	8.627 ± 0.010	6.704 ± 0.026	5.774 ± 0.030	6.239	2.491 ± 0.008
1.01	9.778 ± 0.018	8.826 ± 0.019	6.693 ± 0.078	6.049 ± 0.064	6.371	2.539 ± 0.015
2.02	10.094 ± 0.018	9.117 ± 0.023	7.246 ± 0.045	6.070 ± 0.053	6.658	2.574 ± 0.013
3.09	10.380 ± 0.008	9.379 ± 0.006	7.373 ± 0.046	6.256 ± 0.079	6.815	2.576 ± 0.013
			$CaCl_2$			
0.52	9.493 ± 0.008	8.339 ± 0.011	6.425 ± 0.025	5.518 ± 0.027	5.972	2.459 ± 0.008
1.45	9.832 ± 0.003	8.810 ± 0.004	6.970 ± 0.007	5.808 ± 0.015	6.389	2.583 ± 0.015
2.93	10.076 ± 0.009	9.151 ± 0.011	7.254 ± 0.026	6.127 ± 0.024	6.691	2.681 ± 0.014
5.88	10.369 ± 0.009	9.230 ± 0.021	7.351 ± 0.023	6.219 ± 0.049	6.785	2.783 ± 0.009
			Me_4NCl			
0.55	9.625 ± 0.005	8.539 ± 0.012	6.608 ± 0.039	5.672 ± 0.039	6.140	2.454 ± 0.005
1.09	9.758 ± 0.005	8.711 ± 0.009	6.832 ± 0.015	5.719 ± 0.031	6.275	2.509 ± 0.005
2.39	9.930 ± 0.031	8.880 ± 0.027	6.920 ± 0.067	5.930 ± 0.075	6.430	2.480 ± 0.008
4.06	9.841 ± 0.011	8.898 ± 0.010	7.046 ± 0.017	5.854 ± 0.016	6.450	2.488 ± 0.013

[a] Values calculated from eq 8. [b] Values calculated from eq 9. [c] ± Standard deviation.

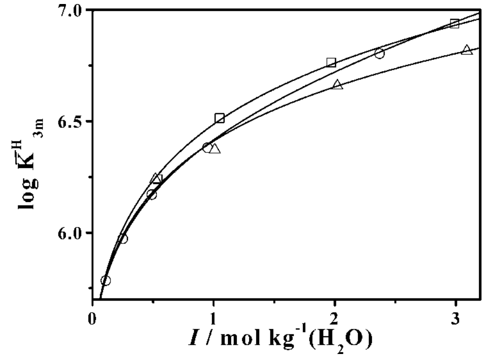

Figure 2. Protonation constant (log $\bar{K}^{H}_{3m}$) at different ionic strengths (I) in □, LiCl; ○, NaCl; and △, KCl, at T = 298.15 K.

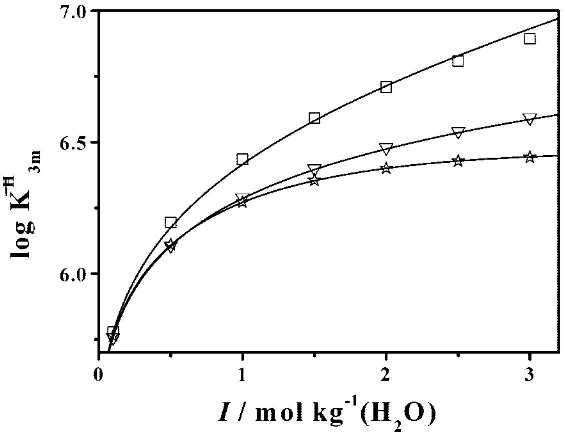

Figure 3. Calculated protonation constant (log $\bar{K}^{H}_{3m}$) at different ionic strengths (I) in □, alkali metal salts; ▽, $CaCl_2$; and ☆, Me_4NCl, at T = 298.15 K.

droxymethyl)amino-methane] with HCl and was estimated to be Q ± 0.008 J and V ± 0.001 mL.

Procedure. Potentiometric Measurements. The titrand solutions were obtained mixing different amounts of BPEI calculated as a monomer (see Table 1), an excess of HCl ($C_{HCl} \approx 5 \times C_{BPEI}$ mmol·L^{-1}) necessary to fully protonate each of the amino groups of BPEI, and the supporting electrolyte to obtain pre-established ionic strength values. Potentiometric measurements were carried out by titrating 25 mL of the titrand solution with standard NaOH solution up to pH ≈ 11. Table 1 shows the experimental conditions. For each titration, 80 to 100 points were collected, and for each experiment, independent titrations of strong acid solution (C_{HCl} = 10 mmol·L^{-1}) with standard base were carried out under the same medium and ionic strength conditions as the systems under study for determining electrode potential (E^0) and acidic junction potential ($E_j = j_a$ [H^+]). For each ionic strength, the potentiometric titrations were carried out in duplicate.

Calorimetric Measurements. Calorimetric measurements were performed by titrating 50 mL of the solution containing BPEI from (1.0 to 2.6) mmol·L^{-1} and the ionic media to obtain the pre-established ionic strength values (0.25 ≤ I/mol·L^{-1} ≤ 5 for LiCl, NaCl, and $CaCl_2$; 0.1 ≤ I/mol·L^{-1} ≤ 3 for KCl) with HCl standard solution. Before each experiment, the heat of dilution was measured under the same ionic strength conditions.

Calculations. The nonlinear least-squares computer program ESAB2M[45] was used for the refinement of all parameters of the acid−base titration (E^0, K_w, analytical concentration of reagents and the protonation degree ($\bar{p}$) of the polyelectrolyte during the titration). The same computer program was also used in the calculation of the conditional protonation constants by minimizing the sum of errors squared on titrant volume at the ionic strength values of each specific titration. The nonlinear least-squares program ES2WC[46] was used to compute the formation constants of weak complexes from potentiometric data. Details of the calculation method and program have already been reported.[47] Calorimetric titration data were analyzed by the ES5CMI[48] computer program. The LIANA[47] computer program was used to fit the potentiometric and calorimetric data to the equations reported in the following sections.

Table 5. Infinite Dilution Protonation Constants of BPEI at T = 298.15 K, Together with $\Delta\varepsilon_\infty$ and $\Delta\varepsilon_0$ Parameters (Molal Concentration Scale)

		$\Delta\varepsilon_\infty$	$\Delta\varepsilon_0$	σ^a
			LiCl	
$\log K^{H0}_1$	9.36^b	0.07 ± 0.03^c	1.03 ± 0.15^c	0.029
$\log K^{H0}_2$	7.90	0.06 ± 0.02	1.17 ± 0.12	0.010
$\log \bar{K}^{H0}_3$	5.29	0.06 ± 0.02	0.70 ± 0.04	0.008
$\log K^{H0}_4$	1.80	0.13 ± 0.01	-0.99 ± 0.16	0.014
			KCl	
		0.22 ± 0.03^c	0.66 ± 0.08^c	0.028
		0.12 ± 0.02	0.91 ± 0.06	0.019
		0.05 ± 0.02	0.56 ± 0.05	0.022
		0.02 ± 0.02	-1.00 ± 0.05	0.015
			$CaCl_2$	
		0.09 ± 0.03^c	0.66 ± 0.15^c	0.037
		0.03 ± 0.03	0.75 ± 0.11	0.080
		0.03 ± 0.02	0.33 ± 0.07	0.066
		0.04 ± 0.03	-0.96 ± 0.11	0.031
			Me_4NCl	
		-0.05 ± 0.02^c	0.84 ± 0.05^c	0.028
		-0.05 ± 0.01	0.82 ± 0.02	0.005
		-0.05 ± 0.01	0.38 ± 0.03	0.008
		0.01 ± 0.01	-1.07 ± 0.04	0.016

[a] Standard deviation on the fit. [b] Infinite dilution values reported ref 35. [c] ± standard deviation.

There are different methods to analyze the potentiometric data of low molecular weight amines and high molecular weight polyamines. By considering the generic reaction

$$AH_{i-1}^{(i-1)+} + H^+ \rightleftarrows AH_i^{i+} \qquad K^H_i \qquad (1)$$

$$A + iH^+ \rightleftarrows AH_i^{i+} \qquad \beta^H_i \qquad (2)$$

(A is the monomeric unit of BPEI and i is the number of protons bound to the amine), the relative protonation constants are not true constants but depend on the protonation degree, $\bar{p}$, defined as

$$\bar{p} = \frac{[A]_T - [A]}{[A]_T} \qquad (3)$$

where $[A]_T$ and [A] are the total and free concentrations of the polyamine, respectively. For a high molecular weight polyamine, there is no single value of the protonation constant since the change on $\bar{p}$ corresponds to the proton–polyamine charge variation. To take into account the effect of $\bar{p}$ on K^H_i, many models have been proposed. One of these is the modified Henderson–Hasselbalch equation[49]

$$\log K^H_i = \log K^H_{ni} - (n_i - 1)\log[(1 - i + \bar{p})/(i - \bar{p})] \qquad (4)$$

This is a two-parameter model, namely $K^H_{n_i}$ (the protonation constant at $i/2$) and n_i, an adjustment parameter which takes into account the deviation from the original Henderson–Hasselbalch equation ($n_i = 1$). A more accurate equation, based on the Guggenheim zeroth approximation,[50] was proposed by Högfeldt[51]

$$\log K^H_i = (i - \bar{p})^2 \log K^H_{i(1)} + (1 - i + \bar{p})^2 \log K^H_{i(0)} + 2(i - \bar{p})(1 - i + \bar{p}) \log K^H_{i(m)} \qquad (5)$$

where $K^H_{(1)}$ and $K^H_{(0)}$ represent the protonation constant when $\bar{p} \to i$ and $\bar{p} \to (i - 1)$, and $K^H_{(m)}$ is an intermediate value which takes into account the nonlinearity of the function log K^H_i vs $\bar{p}$. The thermodynamic protonation constant can be represented by a mean constant ($\bar{K}^H$) obtained by the integral of the experimental curve $\log K^H_i = f(\bar{p})$ in the range $(i - \bar{p}) \le \bar{p} \le (1 - i + \bar{p})$

$$\log \bar{K}^H_i = \int_{i-\bar{p}}^{1-i+\bar{p}} \log K^H_i d\bar{p} \qquad (6)$$

corresponding to the arithmetic mean of the three parameters of eq 5

$$\log \bar{K}^H_i = (\log K^H_{i(1)} + \log K^H_{i(0)} + \log K^H_{i(m)})/3 \qquad (7)$$

If the function log K^H_i vs $\bar{p}$ is linear, eq 5 can be simplified to

$$\log K^H_i = (i - \bar{p}) \log K^H_{i(1)} + (1 - i + \bar{p}) \log K^H_{i(0)} \qquad (8)$$

and therefore

$$\log \bar{K}^H_i = \log K^H_{i(m)} = (\log K^H_{i(1)} + \log K^H_{i(0)})/2 \qquad (9)$$

Equations 4 and 8were used in our studies on BPEI protonation in NaCl aqueous media[35] obtaining a slightly best fit by using eqs 8 and 9.

Results and Discussion

Protonation Constants. By using the modified Henderson–Hasselbalch approach (eq 4), the third protonation step of BPEI is the most dependent on the protonation degree ($\bar{p}$), and this is in agreement with the results previously observed in NaCl.[35] Table 2 shows the results obtained for each supporting electrolyte at $I \approx 0.5$ mol·L^{-1}. To find the best fitting models, three different approaches were applied to the potentiometric data of BPEI. In the first one (m1), the BPEI was considered as a simple low molecular weight tetramine, and each protonation constant was calculated as a true constant; in the second one (m2), the third protonation step was calculated as a linear function of $\bar{p}$ according to eq 8; in the last one (m3), the log K^H_3 was considered as a nonlinear function of $\bar{p}$, by applying eq 5. In Table 3, as an example, the results obtained at $I \approx 0.5$ mol·L^{-1} in each supporting electrolyte are reported, together with the standard deviation in the fit (σ) relative to the fit of the different models and the variance ratio (σ^2/σ_0^2). Model (m2) shows a fairly good improvement on the fit with respect to model (m1), while the difference in σ between (m2) and (m3) models is fairly negligible.

Dependence on Ionic Strength of Protonation Constants. Dependence on ionic strength was taken into account by a Debye–Hückel type equation (at T = 298.15 K)

$$\log K^H_i = \log K^{H0}_i + 2(i - 1)0.51I^{1/2}/(1 + 1.5I^{1/2}) + C_i I \qquad (10)$$

where K^H_i is the formation constant and K^{H0}_i is the formation constant at infinite dilution and C_i is an empirical parameter. When I and K^H_i are expressed in the molal concentration scale, eq 10 becomes the classical and widely used SIT (Specific ion Interaction Theory) equation,[41–43] with $C_i = \Delta\varepsilon_i$ and

$$\Delta\varepsilon_i = \Sigma_j \varepsilon(j,k) \qquad (11)$$

where $\varepsilon(j,k)$ is the SIT interaction coefficient of the jth species (involved in the equilibrium represented by the protonation constant K^H) with the kth component of opposite charge. The specific interaction coefficients $\varepsilon(j,k)$ are, in the original model, true constants, but this approximation is valid only for some electrolytes and for some ionic strength ranges. In the modified version of the SIT approach,[52–54] these coef-

Table 6. Protonation constants of BPEI in different salts calculated from eq 10, at different ionic strengths in the molal concentration scale, at $T = 298.15$ K

I /mol·kg^{-1}		LiCl	NaCl[a]	KCl	alkali metal salts[b]	$CaCl_2$	Me_4NCl
0.1	$\log K^{H}_{1}$	9.454	9.451	9.422	9.442 ± 0.018	9.421	9.436
	$\log K^{H}_{2}$	8.225	8.214	8.203	8.214 ± 0.011	8.187	8.193
	$\log \bar{K}^{H}_{3}$[c]	5.792	5.768	5.779	5.780 ± 0.012	5.758	5.762
	$\log K^{H}_{4}$	2.368	2.393	2.366	2.376 ± 0.015	2.370	2.359
1	$\log K^{H}_{1}$	9.911	9.942	9.801	9.885 ± 0.074	9.733	9.751
	$\log K^{H}_{2}$	8.920	8.906	8.826	8.884 ± 0.051	8.701	8.693
	$\log \bar{K}^{H}_{3}$[c]	6.488	6.410	6.410	6.436 ± 0.045	6.289	6.272
	$\log K^{H}_{4}$	2.597	2.693	2.536	2.609 ± 0.079	2.566	2.491
2	$\log K^{H}_{1}$	10.141	10.271	10.096	10.169 ± 0.091	9.918	9.845
	$\log K^{H}_{2}$	9.217	9.265	9.135	9.206 ± 0.066	8.909	8.841
	$\log \bar{K}^{H}_{3}$[c]	6.766	6.714	6.653	6.711 ± 0.057	6.480	6.401
	$\log K^{H}_{4}$	2.707	2.776	2.553	2.679 ± 0.114	2.603	2.479
2.5	$\log K^{H}_{1}$	10.222	10.409	10.227	10.286 ± 0.107	9.990	9.861
	$\log K^{H}_{2}$	9.315	9.402	9.251	9.323 ± 0.076	8.977	8.872
	$\log \bar{K}^{H}_{3}$[c]	6.861	6.831	6.735	6.809 ± 0.066	6.542	6.428
	$\log K^{H}_{4}$	2.769	2.812	2.566	2.716 ± 0.131	2.625	2.479
3	$\log K^{H}_{1}$	10.292	10.538	10.353	10.394 ± 0.128	10.056	9.865
	$\log K^{H}_{2}$	9.397	9.525	9.354	9.425 ± 0.089	9.032	8.890
	$\log \bar{K}^{H}_{3}$[c]	6.941	6.938	6.804	6.894 ± 0.078	6.594	6.443
	$\log K^{H}_{4}$	2.834	2.846	2.580	2.753 ± 0.150	2.648	2.481

[a] Ref 35. [b] Mean value ± mean deviation for alkali metal salts. [c] Values obtained from eq 7.

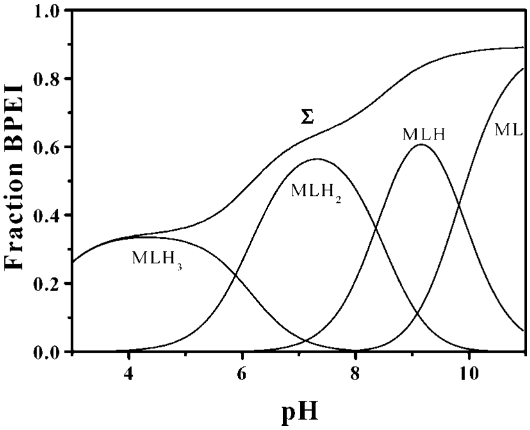

Figure 4. Fraction of complexed BPEI in the presence of Ca^{2+} at $T = 298.15$ K and $I = 1.0$ mol·kg^{-1}. Analytical conditions: $C_{Ca} = 0.333$ mol·kg^{-1}; $C_{BPEI} = 1$ mmol·kg^{-1} (M = Ca^{2+}; L = BPEI). Charge omitted for simplicity.

ficients and the relative differences $\Delta\varepsilon_i$ are expressed as a function of I

$$\Delta\varepsilon_i = \Delta\varepsilon_{\infty i} + \frac{\Delta\varepsilon_{0i} - \Delta\varepsilon_{\infty i}}{I + 1} \quad (12)$$

where $\Delta\varepsilon_i = \Delta\varepsilon_{\infty i}$ for $I \to \infty$ and $\Delta\varepsilon_i = \Delta\varepsilon_{0i}$ for $I \to 0$. To apply the SIT approach, the molar protonation constants were converted to the molal concentration scale following a procedure already described,[55] and the molal protonation constants are reported in Table 4. $\Delta\varepsilon_i$ values are reported in Table 5 (using the infinite dilution values, $\log K^{H}{}_i$ reported in ref 35). Differences between the protonation constants in different alkali metal salts (see Figure 2) are quite small, and to simplify the comparison between the different ionic media, the mean value of protonation constants in alkali metal salt solutions were calculated (see Table 6). Its dependence on ionic strength, by applying eqs 10 and 12, is reported in Table 7. By analyzing the data in Table 6, the decrease of protonation constant values in Ca^{2+} and Me_4N^+ with respect to the values in alkali metal salts is evident. As an example, the trend of $\log \bar{K}^{H}{}_3$ vs ionic strength, for different ionic media, is shown in Figure 3.

Table 7. Dependence on Ionic Strength Parameters of BPEI Referred to Molal Constants at $T = 298.15$ K in Alkali Ionic Media

		alkali metal ionic media		
		$\Delta\varepsilon_\infty$	$\Delta\varepsilon_0$	σ[a]
$\log K^{H}_{1}$	9.36[b]	0.20 ± 0.03[c]	0.84 ± 0.07[c]	0.079
$\log K^{H}_{2}$	7.90	0.14 ± 0.02	0.98 ± 0.05	0.053
$\log \bar{K}^{H}_{3}$	5.29	0.13 ± 0.02	0.49 ± 0.04	0.038
$\log K^{H0}_{4}$	1.80	0.10 ± 0.01	−0.95 ± 0.03	0.065

[a] Standard deviation on the fit. [b] Protonation constants at infinite dilution reported in ref 35. [c] ± standard deviation.

Calcium and Me_4N^+ Complexes. The lowering of protonation constants in $CaCl_2$ and Me_4NCl can be explained by considering the formation of weak complexes between Ca^{2+} or Me_4N^+ and unprotonated or partially protonated BPEI. Our calculations are based on the comparison of different protonation constants obtained in the two background salts at different concentrations and ionic strengths and assuming no interactions between alkali metal ions with unprotonated or partially protonated BPEI. Also, the interactions of Cl^- and $(BPEI)H_i^{i+}$ are not considered.[56] The average number of protons bound to the ligand ($\bar{p}$, $\bar{p}^*$) is defined by the equation

$$\bar{p}^* = \frac{\Sigma i\beta_i^{H*}[H]^i}{1 + \Sigma\beta_i^{H*}[H]^i} \quad (13)$$

where β_i^{H*} is the overall protonation constant; $\beta_i^{H*} = \Pi K_i^{H*}$; and the superscript * indicates a conditional constant. If the baseline K_i^{H*} values are known (and in own case K^{H} and K^{H*} are the protonation constants obtained in solutions containing not interacting and interacting components, respectively), eq 13 can be also written

$$\bar{p} = \frac{\sum i\beta_i^{H}[H]^i(1 + K_i^{M}[M])}{1 + \sum \beta_i^{H}[H]^i(1 + K_i^{M}[M])} \quad (14)$$

If the assumptions (a) the activity coefficients of the baseline solution and that containing the interaction cation are, within acceptable errors, constant and (b) the stability of weak complexes is sufficiently low and the concentration of interacting cation(s) is sufficiently high to make the approximation [M] = total concentration of M, can be

Table 8. Complex Formation Constants in Ca^{2+}−BPEI and Me_4N^+−BPEI Systems, at T = 298.15 K and I = 1.0 mol·kg^{-1}

	$CaCl_2$	Me_4NCl
K_{10}[a]	24.9 ± 1.7[b]	24 ± 10[b]
K_{11}	19 ± 2	10 ± 4
K_{12}	5.4 ± 0.7	3.0 ± 1.2
K_{13}	1.6 ± 0.5	1.0 ± 0.4

[a] Referred to reaction (iM + H_jBPEI = M_iH_jBPEI; K_{ij}) [charge omitted for simplicity]. [b] ± standard deviation.

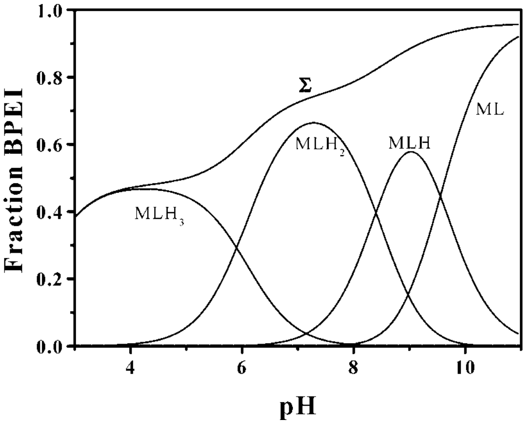

Figure 5. Fraction of complexed BPEI in the presence of Me_4N^+ at T = 298.15 K and I = 1.0 mol·kg^{-1}. Analytical conditions: $C_{Me_4}N$ = 1.0 mol·kg^{-1}; C_{BPEI} = 1 mmol·kg^{-1} (M = Me_4N^+; L = BPEI). Charge omitted for simplicity.

considered correct, the values of K^{H}_{i} can be calculated by minimizing the deviations square sum

$$\sum (p - p^{*})^2 \qquad (15)$$

Least squares analysis[46] allowed us to calculate the stability of four species for both the Ca^{2+}−BPEI and Me_4N^+−BPEI system

$$CaH_iBPEI^{(2+i)} \quad (i = 0,1,2,3)$$

$$(Me_4N)H_iBPEI^{(1+i)} \quad (i = 0,1,2,3)$$

relative to the equilibria

$$Ca^{2+} + H_iBPEI^{i+} = CaH_iBPEI^{(2+i)} \qquad K^{Ca}_{i} \qquad (16)$$

$$Me_4N^{+} + H_iBPEI^{i+} = (Me_4N)H_iBPEI^{(1+i)} \qquad K^{Me_4N}_{i} \qquad (17)$$

This type of calculation is correct only for moderate ionic strengths [(0 ≤ I ≤ 1) mol·kg^{-1}]. Table 8 reports the values of formation constants (at T = 298.15 K, I = 1.0 mol·kg^{-1}), and in Figures 4 and 5, the speciation diagrams of BPEI in $CaCl_2$ and Me_4NCl at the same ionic strength are shown. In these conditions, the binding capacity of BPEI toward the two cations is fairly similar, in the whole pH range 2 to 11, with a small trend $Me_4N^+ < Ca^{2+}$. As expected, the percentage of complexed BPEI increases with increasing pH.

Protonation Enthalpies and Their Dependence on Ionic Strength. The results of calorimetric measurements are shown in Table 9 where the enthalpies and the entropies for the overall protonation equilibria of BPEI at different ionic strengths are reported together with the corresponding ΔG values calculated from the protonation constants in each media converted in the molal concentration scale. Because

Table 9. ΔG, ΔH, and TΔS for the Overall Protonation Equilibria of BPEI at Different Ionic Media and Ionic Strengths, at T = 298.15 K

I(LiCl) mol·kg^{-1}	i[a]	−ΔG_i kJ·mol^{-1}	ΔH_i kJ·mol^{-1}	TΔS_i kJ·mol^{-1}	I(NaCl) mol·kg^{-1}	i[a]	−ΔG_i kJ·mol^{-1}	ΔH_i kJ·mol^{-1}	TΔS_i kJ·mol^{-1}
0.25	1	54.6	−47.8	6.8	0.25	1	54.6	−47.8	6.8
	2	102.7	−92.7	10.0		2	102.6	−92.7	9.9
	3	137.0	−122.5	14.5		3	136.7	−122.5	14.2
	4	151.1	−140.4	10.7		4	151.2	−139.0	12.2
1.02	1	56.5	−48.2	8.3	1.03	1	56.8	−46.6	10.2
	2	107.4	−92.6	14.8		2	107.7	−91.7	16
	3	144.4	−122	22.4		3	144.3	−121.9	22.4
	4	159.2	−136.2	23.0		4	159.7	−136.3	23.4
3.2	1	58.7	−50.6	8.1	3.21	1	60.6	−44.0	16.6
	2	112.3	−94.5	17.8		2	115.4	−89.5	25.9
	3	151.9	−125.2	26.7		3	155.4	−121.9	33.5
	4	168.0	−132.2	35.8		4	171.9	−133.2	38.7
5.59	1	60.0	−52.1	7.9	5.63	1	63.4	−41.1	22.3
	2	115.0	−98.1	16.9		2	120.7	−89.4	31.3
	3	156.0	−133.1	22.9		3	162.9	−125.3	37.6
	4	173.7	−136.9	36.8		4	180.2	−130.2	50.0

I(KCl) mol·kg^{-1}	i[a]	−ΔG_i kJ·mol^{-1}	ΔH_i kJ·mol^{-1}	TΔS_i kJ·mol^{-1}	I($CaCl_2$) mol·kg^{-1}	i[a]	−ΔG_i kJ·mol^{-1}	ΔH_i kJ·mol^{-1}	TΔS_i kJ·mol^{-1}
0.1	1	53.9	−47.5	6.4	0.24	1	54.1	−48.6	5.5
	2	100.8	−92.2	8.6		2	101.6	−92.9	8.7
	3	133.8	−121.4	12.4		3	135.4	−120.1	15.3
	4	147.3	−138.3	9.0		4	149.5	−132.9	16.6
0.25	1	54.6	−46.9	7.7	0.99	1	55.5	−51.0	4.5
	2	102.7	−90.9	11.8		2	105.1	−94.9	10.2
	3	137.0	−119.1	17.9		3	141.0	−122.5	18.5
	4	151.1	−136.5	14.6		4	155.7	−132.4	23.3
1.03	1	56.6	−45.2	11.4	3.06	1	57.5	−56.3	1.2
	2	107.6	−88.3	19.3		2	109.1	−101.0	8.1
	3	144.7	−114.9	29.8		3	146.8	−129.6	17.2
	4	159.6	−133.2	26.4		4	162.0	−134.6	27.4
3.31	1	59.0	−39.8	19.2	5.21	1	58.9	−61.4	−2.5
	2	112.9	−80.9	32.0		2	111.6	−107.4	4.2
	3	152.8	−104.8	48.0		3	150.3	−137.6	12.7
	4	169.3	−127.8	41.5		4	166.2	−138.3	27.9

[a] Overall protonation: BPEI + $i$$H^+$ ⇄ (BPEI) H_i^{i+}.

Table 10. Infinite Dilution Values of $\Delta H^0{}_i$ (at $I = 0$ mol·kg^{-1}) of BPEI at $T = 298.15$ K Together with $\Delta\varepsilon'$ Parameters (Equation 18)

		$\Delta\varepsilon'$ (LiCl)	$\Delta\varepsilon'$ (NaCl)	$\Delta\varepsilon'$ (KCl)	$\Delta\varepsilon'$ ($CaCl_2$)
$\Delta H^0{}_1$	−47.84 ± 0.07[a]	−0.77 ± 0.04[a]	1.19 ± 0.01[a]	2.44 ± 0.08[a]	−2.65 ± 0.09[a]
$\Delta H^0{}_2$	−45.23 ± 0.07	−0.36 ± 0.07	−0.78 ± 0.08	0.82 ± 0.02	−0.4 ± 0.05
$\Delta H^0{}_3$	−30.05 ± 0.13	−1.41 ± 0.05	−1.62 ± 0.08	0.96 ± 0.05	−0.56 ± 0.08
$\Delta H^0{}_4$	−18.93 ± 0.15	1.88 ± 0.02	1.52 ± 0.05	−2.57 ± 0.10	2.82 ± 0.07

[a] Standard deviation.

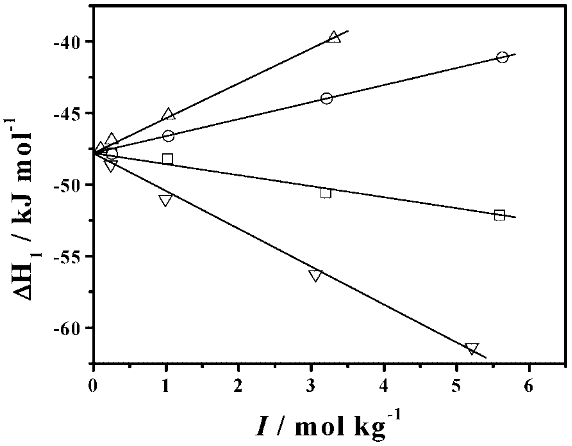

Figure 6. ΔH_1 values of BPEI vs ionic strength (I) in △, KCl; ○, NaCl; □, LiCl; and ▽, $CaCl_2$ at $T = 298.15$ K.

of the shameful muddle in our computer files, the enthalpy changes for the protonation of BPEI in NaCl shown in our previous paper[35] are incorrect; the correct ones are reported in Table 9 of this paper. As can be observed from this table and from Figure 6 (where, as an example, the ΔH_1 vs the ionic strength variation is reported), the ΔH for low ionic strengths are quite similar, whereas for higher ionic strengths, the differences are evident and the trend is KCl > NaCl > LiCl > $CaCl_2$ for each protonation step.

The dependence of protonation enthalpy ΔH_i on the ionic strength can be expressed by the equation

$$\Delta H_i = \Delta H^0{}_i + A'2(i-1)\sqrt{I}/(1+1.5\sqrt{I}) + I\Delta\varepsilon'_i \qquad (18)$$

where $\Delta H^0{}_i$ is the enthalpy change at $I \rightarrow 0$ referred to each protonation step, $A' = RT^2 \ln 10\,(\mathrm{d}A/\mathrm{d}T) = 1.5$, and

$$\Delta\varepsilon'_i = RT^2 \ln 10 \frac{\mathrm{d}\Delta\varepsilon}{\mathrm{d}T}$$

In Table 10, protonation enthalpies of BPEI at infinite dilution together with parameters for the dependence on ionic strength are reported.

Conclusions

The protonation behavior of BPEI is very similar to that of low molecular weight polyamines as evidenced by the following considerations.

Trends of protonation constants for several low molecular weight polyamines were discussed in refs 57 and 58 as a function of different factors, such as the length of the alkyl chain and the presence of primary, secondary, or tertiary amino groups. Values of protonation constants found for BPEI are consistent with those of low molecular weight amines [in particular with the tetramine-tris(2-aminoethyl)amine].

Very few studies were published on the effect of different salts on acid−base properties of polyamines,[36,38,59,60] and the trend for log K^H was always NaCl ≈ KCl. Recently, protonation constants of a high molecular weight polyamine, namely, polyallylamine,[38] were determined in LiCl and NaCl aqueous solutions from (0 to 5) mol·L^{-1}. In this case, a small trend $Na^+ > K^+$ was observed, such as in the case of BPEI studied here.

Calcium complexes of polyamines are quite weak.[56,61] Formation constants of dipentamines range between (1.5 to 71) kg·mol^{-1} at $I = 0$ and $T = 298.15$ K. For the Ca(BPEI) complex species, we calculated $K = 25$ kg·mol^{-1} (at $I = 1$ kg·mol^{-1}, $T = 298.15$ K) which is comparable with the above values if the different ionic strength conditions and the large errors associated with such small formation constants are taken into account. Small formation constants were found for low molecular weight polyamine[36,37] complexes with Me_4N^+. Also in this case, as for calcium complexes, the values here found for BPEI can be considered, as a first approximation, quite similar to those of simple amines. The model which uses the formation of alkali metal ion pairs to explain dependence on medium suffers from two weaknesses: (i) the arbitrary choice of a baseline background electrolyte and (ii) the implicit assumption that $\gamma_A = \gamma_{A'}$ and $\gamma_{HA} = \gamma_{HA'}$, where γ_A and γ_{HA} are the activity coefficients of A and HA in the baseline electrolyte, and $\gamma_{A'}$ and $\gamma_{HA'}$ are the same quantities in the interacting electrolyte, corrected for ion pair formation. The first issue has been discussed in many papers and recently in a review.[56] Moreover, two studies of weak complexes using the differences in protonation constants (eqs 13 to 15), often called the Δ log K^H method, and some ISE electrodes[62,63] showed that consistent results are obtained in the calculation of small formation constants. For weak amine interactions, it would seem reasonable to assume NaCl to be the baseline, though not an absolute one. The second assumption is more difficult to justify, but here again empirical evidence is of help. First of all, it is necessary to define an ionic strength range in which the assumption may be valid, and this was indicated in previous works[56,62,63] to be $0 \leq I(\mathrm{mol \cdot L^{-1}}) \leq 1$. In this range, the results obtained using Δ log K^H and ISE methods are quite consistent. Even if these factors do not unambiguously demonstrate the validity of the method, they furnish two important pieces of information, namely, an estimate of the formation constants of amine weak metal complexes and a comparison of the strengths of different weak complexes for different metal−N−ligand systems. In particular, the last point must be affected by the above assumptions to a lesser extent.

Protonation enthalpies of BPEI are higher than those of unsubstituted low molecular weight polyamines and lower with respect to those of substituted ones.[38,57] This is due to a significant entropy contribution in the protonation of both substituted simple amines and BPEI. This behavior is not observed for polyallylamine where only primary amino groups are involved in the protonation.

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