

Synthesis and Protonation Behavior of Carboxylate-Functionalized Poly(propyleneimine) Dendrimers

R. C. van Duijvenbode, A. Rajanayagam, and G. J. M. Koper*

Leiden Institute of Chemistry, Gorlaeus Laboratories, Leiden University, P.O. Box 9502, 2300 RA Leiden, The Netherlands

M. W. P. L. Baars, B. F. M. de Waal, and E. W. Meijer

Laboratory of Macromolecular and Organic Chemistry, Eindhoven University of Technology, P.O. Box 513, 5600 MB Eindhoven, The Netherlands

M. Borkovec

Department of Chemistry, Clarkson University, Potsdam, New York 13699-5814

Received June 18, 1999; Revised Manuscript Received October 19, 1999

ABSTRACT: Five generations of carboxylate-functionalized poly(propyleneimine) dendrimers have been synthesized starting from a double Michael addition of amine-functionalized poly(propyleneimine) dendrimers to methyl acrylate followed by basic hydrolysis using LiOH in a water/methanol mixture. The dendritic compounds have been characterized using ^1H NMR, ^{13}C NMR, IR, and ESI–MS spectroscopy. Subsequently, the protonation behavior of these potential complexing agents is studied with potentiometric titrations in 0.1 and 1.0 M KCl solutions. The carboxylate-functionalized dendrimers show a characteristic onionlike shell protonation behavior. The titration curves for different ionic strengths cross at pH 8.5 which is exactly after protonation of the outermost shell of amines ($\theta \sim 0.25$). This crossing point is found not to be identical to the point of zero charge $\theta \sim 0.5$, which is observed at pH 4.5. The Ising model was used to rationalize the complete set of titration data simultaneously for five generations with only one microscopic pK value for each additional shell and a set of three nearest neighbor pair interaction parameters.

Introduction

In the past decade dendritic molecules^{1–3} have attracted an enormous amount of attention and successfully proven to be a new subclass of molecules between the low molecular organic compounds and the conventional high polymeric structures.

The physicochemical properties of dendrimers are sometimes in sharp contrast to the behavior of linear polymers because of their extraordinary symmetry and high density of terminal functionalities. One example is the specific viscosity, which for the dendrimers or starburst polymers goes through a maximum as a function of molecular weight, where linear polymers show a monotonic increase. This maximum in the viscosity is directly related to the maximum in the specific volume of the molecules. Their mass increases by a factor of 2 per generation whereas the radius of the molecules increases linearly with generation and hence the specific volume varies as $n^3 2^{-n}$ with generation n .

Specific chemical features of dendritic molecules can be obtained by introducing the desired functional groups at the periphery or by using a functional skeleton with coordination sites for specific interactions.⁴ Acid-functionalized poly(propyleneimine) dendrimers not only consist of coordination sites at the periphery but also contain a functional interior of tertiary amines. These compounds have been used previously in electrokinetic chromatography.⁵ The synthetic route in this case

involved acidic hydrolysis of the nitrile-functionalized poly(propyleneimine) dendrimers,⁶ yielding carboxylic acid-functionalized dendrimers. These synthesized compounds contain a high concentration of salts (~ 30 wt %), are very hygroscopic, and have a brownish color, in particular for the higher generations. Therefore, a new route toward acid-functionalized poly(propyleneimine) dendrimers was developed starting from the commercially available amine-functionalized poly(propyleneimine) dendrimers,⁶ i.e. DAB-*dendr*-(NH_2) _{$n/2$} ($n = 4, 8, 16, 32, 64$).⁷ Michael addition of 2 equiv of methyl acrylate to the amines of the dendrimer yields methyl ester-functionalized poly(propyleneimine) dendrimers, i.e. DAB-*dendr*-(COOMe) _{n} ($n = 4–64$). Basic hydrolysis using LiOH in a water/methanol mixture yields carboxylate-functionalized dendrimers, denoted as DAB-*dendr*-(COO[–]Li⁺) _{n} ($n = 4, \mathbf{1}; n = 8, \mathbf{2}; n = 16, \mathbf{3}; n = 32, \mathbf{4}; n = 64, \mathbf{5}$) as white powders. The synthesis is outlined in Figure 1. ESI–MS spectra of generations $\mathbf{1}–\mathbf{4}$ are presented in Figure 2. Up to generation $\mathbf{4}$ (almost) no defect structures are observed, indicating that the purity of the compounds is very high. ESI–MS analysis of generation $\mathbf{5}$ proved unsuccessful under the used conditions, however this is not due to an ill-defined structure as can be derived from other characterization techniques (^1H NMR, ^{13}C NMR, IR). The position of the parent peaks can be assigned to the theoretically predicted mass; no defect structures are observed.

The first generation, i.e. DAB-*dendr*-(COO[–]Li⁺)₄ ($\mathbf{1}$), closely resembles EDTA, which contains the same two inner amines and four outer carboxylic groups, albeit

* To whom correspondence should be addressed.

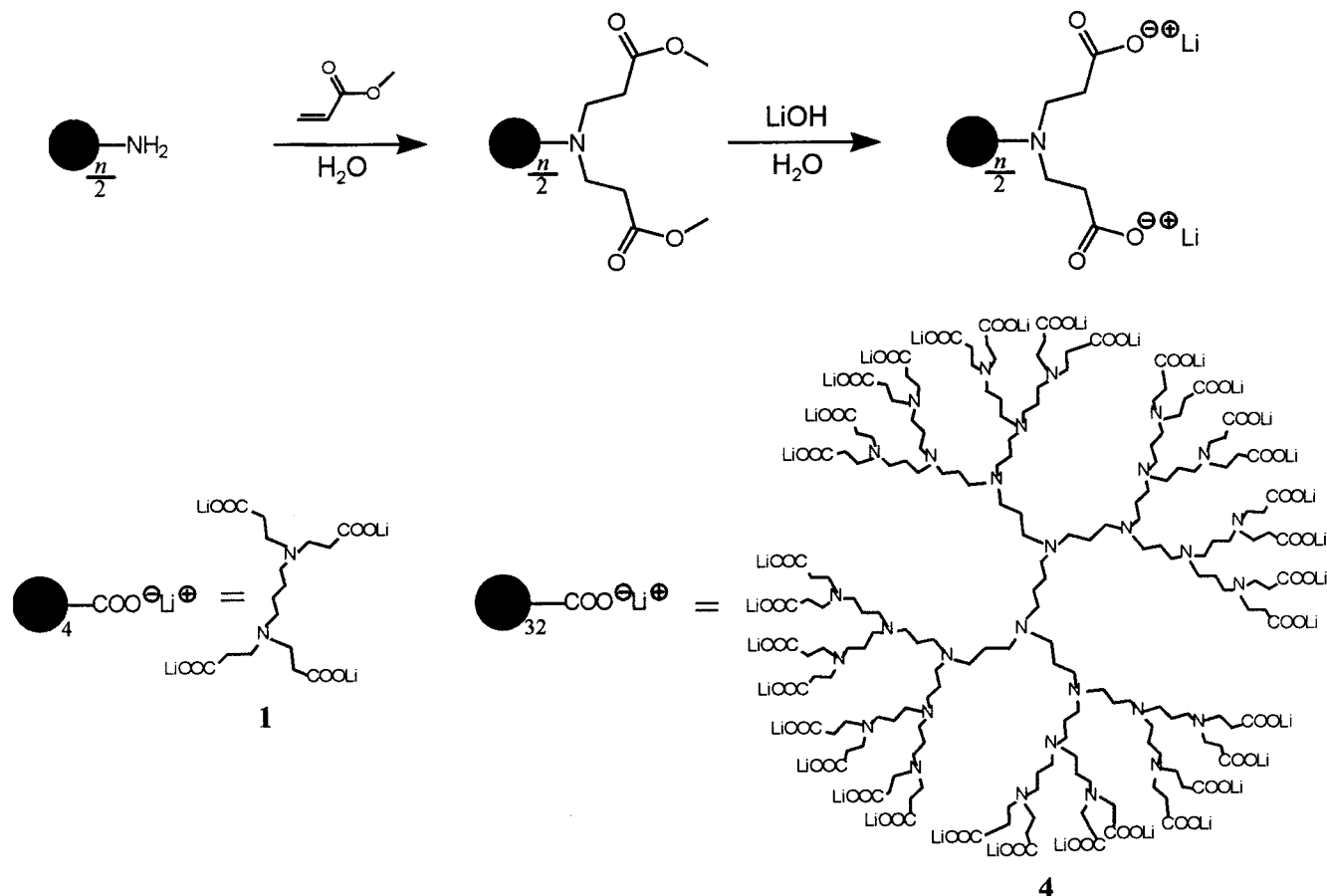


Figure 1. Synthetic scheme toward carboxylate-functionalized poly(propyleneimine) dendrimers **1**–**5**. Only the periphery is drawn; the core and the interior are indicated with the black circles. The chemical structure of generations **1** and **4** are depicted in detail. n represents the number of terminal carboxylate functionalities (4, 8, 16, 32, and 64 respectively for generations **1**–**5**). Thus, $n/2$ is the number of amine-end groups of the amine-functionalized poly(propyleneimine) dendrimers used as a reagent in the synthesis.

separated by a smaller spacer. EDTA is well-known for its complexing power. All generations of the carboxylate-functionalized poly(propyleneimine) dendrimers consist of similar structural elements but contain considerably more coordination sites than EDTA. Because of the high local concentration of functional groups, these treelike molecules possess a very high charge density. The charge density and the functional interior make them potential complexing agents,⁸ which would appropriately be called polydentate ligands.

As a first step in understanding the complexing power of these modified poly(propyleneimine) dendrimers, in the present article the acid–base properties of these molecules will be studied. The interpretation of the titration data is based on the Ising model, which has been shown in the past to be a quantitative tool to study the acid–base properties of various linear polyelectrolytes with one type of ionizable group.^{9–12} The Ising model parametrizes the protonation behavior of the polyelectrolyte with one microscopic pK value (given that all other groups in the molecule are deprotonated) for each type of protonable group and interaction energies for pairs and possibly triplets of protonated sites. The Ising model was applied successfully to polyelectrolytes such as linear poly(ethylene imine)¹³ and poly(carboxylic acids).^{11,12,14} The linear polyelectrolytes all show, some more distinctly than others, a two-step protonation behavior with a plateau at degree of protonation $\theta = 1/2$. This plateau is related to a stable intermediate protonation state with alternating proto-

nated and deprotonated groups along the chain, which is due to the largely pairwise interaction between neighboring protonated groups.

The method has previously been used to explain the protonation behavior of the amine-functionalized poly(propyleneimine) dendrimers.¹⁵ These molecules also exhibit a two-step protonation behavior like the linear polyelectrolytes, albeit with a plateau at a degree of protonation of $2/3$ instead of $1/2$. This plateau corresponds to a similar intermediate state with every second shell protonated, starting with a protonated outer shell of primary amines. This onionlike shell protonation behavior is demonstrated more clearly in ¹⁵N NMR titrations.¹⁶

In the case of the carboxylate-functionalized poly(propyleneimine) dendrimers the Ising model needs to be extended to a branched polyelectrolyte with a combination of two types of ionizable groups. The different groups give rise to an overall charge that varies from negative at high pH values to positive at low pH values.

In the present article, we shall demonstrate that for dendritic molecules with various types of ionizable groups the protonation behavior can perfectly be described in terms of an Ising model. The same onionlike shell protonation behavior emerges. With only a few microscopic pK values, one for each additional shell (with increasing generation), and three nearest neighbor pair interaction parameters, the titration curves of five generations of carboxylate-functionalized dendrimers are simultaneously rationalized.

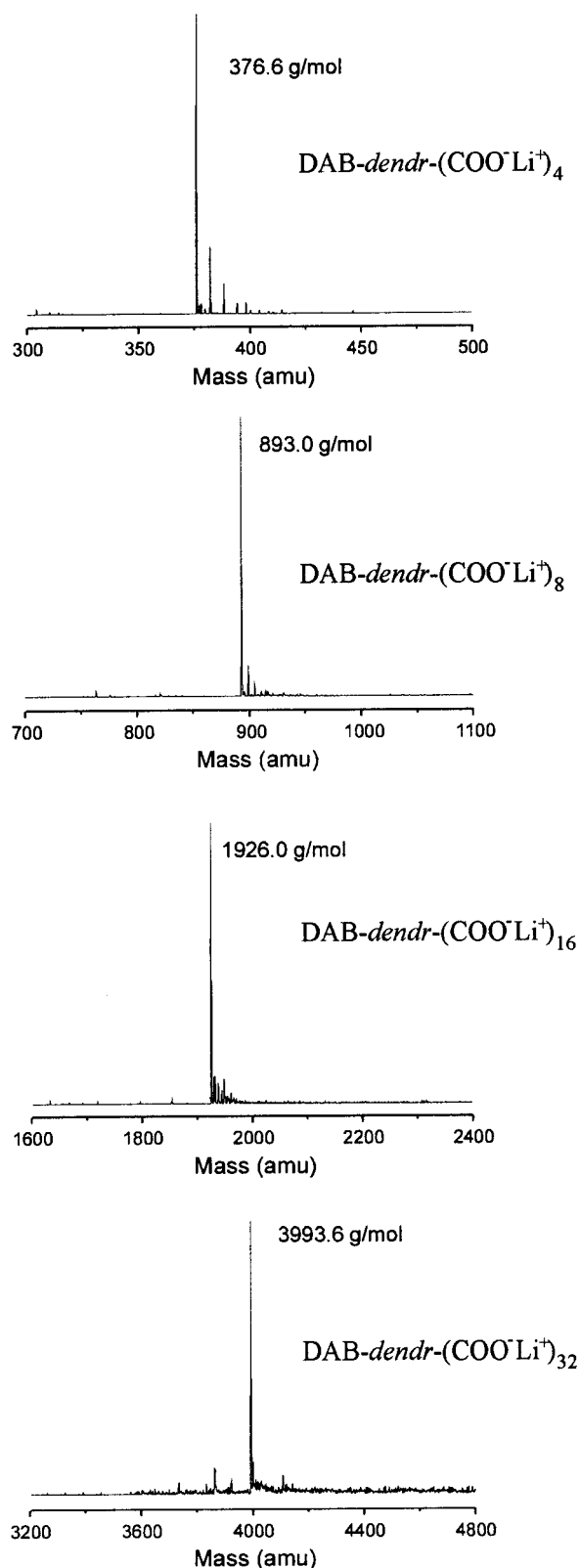


Figure 2. ESI-MS of carboxylate dendrimers 1–4. Parent peaks can be assigned to the correct products. Up to dendrimer 4 almost no defect structures are observed. ESI-MS analysis of generation 5 proved unsuccessful under the conditions used; however, this is not due to an ill-defined structure as can be derived from the other characterization techniques. The small peaks at higher mass can be attributed to (multiple) Li⁺ ions, which coordinate to the carboxylate moiety and originate from the synthetic procedure.

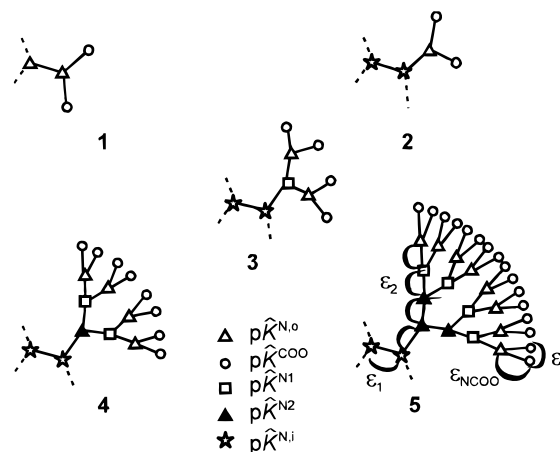


Figure 3. Schematic representation of the dendrimers with an explanation of the parameters of the Ising model used to fit the titration curves simultaneously. The $p\hat{K}_i$ parameters are indicated by the shape of the symbol; the pair interactions ϵ_{ij} are shown for the highest generation.

Ising Model

The Ising model has been used quite extensively throughout the literature to describe the acid–base properties of various polyelectrolytes with a uniform type of ionizable sites.^{9,10,13,17} The approach can be extended to molecules with different ionizable sites, such as primary, secondary, and tertiary amine groups in polyamines¹⁸ and various amino acids in proteins^{19–21} and, as discussed here, to carboxylate-functionalized poly(propyleneimine) dendrimers.

A particular protonation state of a molecule is characterized by a set of N state variables $\{s_1, \dots, s_N\}$, where N is the total number of ionizable sites. The protonation state of a site can be described with $s_i = 0$ if deprotonated and $s_i = 1$ if protonated. Under rather general conditions, the free energy F of formation of a given protonation state $\{s_1, \dots, s_N\}$ can be expanded as

$$\frac{F(s_1, \dots, s_N)}{kT \ln 10} = \sum_i (\text{pH} - p\hat{K}_i) s_i + \frac{1}{2} \sum_{i \neq j} \epsilon_{ij} s_i s_j + \dots \quad (1)$$

where kT denotes the thermal energy, and pH is the common negative logarithm of the proton activity. $p\hat{K}_i$, the logarithm of the dissociation constant for site i , reflects the energy needed to protonate site i considering all other groups deprotonated. Higher order terms other than the pair interactions ϵ_{ij} between two protonated sites are usually left out under the assumption of short-ranged electrostatic interactions. Calculations are further simplified by taking only nearest neighbor pair interactions into account, which for polyamines has proven to cover the larger part of the interactions (largely of electrostatic origin) along the chain.²² The titration curves are readily derived from the above free energy.¹⁸

Using a trivial extension of the recursion method discussed before¹⁸ the Ising model can be evaluated to predict the protonation behavior of the five generations of the present dendrimers simultaneously with the parameters described below. The sites in the different shells of the carboxylate-functionalized poly(propyleneimine) dendrimers are parametrized with microscopic $p\hat{K}_i$ values as indicated in Figure 3. Only the extra inner amine-shells of the two highest generations can be parametrized with the same $p\hat{K}^{(N2)}$ because the chemical

environment is more or less similar. The pair interaction parameter ϵ_{ij} between neighboring amines depends on the spacer length. The ϵ_1 covers the interactions within the diaminobutane core with four carbon atoms between the two amines; for all propyleneimine segments with three carbon spacers, ϵ_2 is used. The Coulombic interactions between the amines and carboxylic groups are reflected by ϵ_{NCOO} . To describe the protonation behavior of poly(carboxylic acids), such as poly(maleic acid), where interactions between neighboring carboxylic groups are rather strong, possibly due to conformation effects,¹⁴ another parameter is used to describe the interactions. This rationalizes the parameter ϵ' of the outer carboxylic groups in the dendritic compounds.

Experimental Section

Materials. Methanol (p.a., Biosolve), toluene (p.a., Biosolve), LiCl (98%, Merck), LiOH·H₂O (99%, Acros Chimica), methyl acrylate (99%, Merck), and hydrochloric acid (37+ %, Vel) were used as received. Water was deionized before use. Amine-functionalized poly(propyleneimine) dendrimers, i.e., DAB-dendr-(NH₂)_{n/2} ($n = 4, 8, 16, 32$ and 64), were obtained from DSM Research, Geleen, The Netherlands. All dendrimers were stripped twice with toluene before use, except for 1,4-diaminobutane, which was distilled prior to use.

Typical Synthesis Procedure for DAB-dendr-(COO⁻Li⁺)₃₂ (4). To a stirred and cooled (ice/water) solution of DAB-dendr-(NH₂)₁₆ (2.02 g, 1.19 mmol) and LiCl (12 mg) in methanol (14 mL) was added methyl acrylate (4.95 g, 57.48 mmol) in a dropwise fashion during 10 min. After the ice/water bath was removed, the reaction mixture was stirred overnight at room temperature and the solvent was evaporated in vacuo. The residue was stripped with dichloromethane in order to remove excess methanol and methyl acrylate, yielding the intermediate methyl ester functionalized dendrimer, DAB-dendr-(COOMe)₃₂ (5.21 g, 98%) as a colorless oil.

¹H NMR (CDCl₃): $\delta = 3.65$ (s, 96H, COOCH₃), 2.75 (t, $J = 7.2$ Hz, 64H, NCH₂CH₂COOCH₃), 2.43 (t, $J = 7.1$ Hz, 64H, NCH₂CH₂COOCH₃), 2.40 (m, 116H, NCH₂CH₂CH₂N {112H} + NCH₂CH₂CH₂CH₂N {4H}), 1.56 {m, 56H, NCH₂CH₂CH₂N}, 1.39 (m, 4H, NCH₂CH₂CH₂CH₂N).

LiOH·H₂O (0.931 g, 22.2 mmol) was added to a stirred and cooled (ice/water bath) solution of DAB-dendr-(COOMe)₃₂ (3.08 g, 0.69 mmol) in methanol (10 mL) and water (6 mL). After the ice/water bath was removed, the reaction mixture was stirred overnight and the solvent was evaporated in vacuo. The remaining oil was redissolved in methanol and again evaporated in vacuo. Drying under vacuum resulted in 2.84 g (98%) of a white powder 4, denoted as DAB-dendr-(COO⁻Li⁺)₃₂.

¹³C NMR (D₂O): $\delta = 184.2$ (32C, COOH), 56.2 (2C, NCH₂CH₂CH₂N), 54.3 (4C, NCH₂CH₂CH₂N), 53.6/53.4 (52C, NCH₂CH₂CH₂N {4C} + NCH₂CH₂CH₂N {16C} + NCH₂CH₂CH₂N {32C}), 52.1 (32C, NCH₂CH₂COOH), 36.7 (32C, NCH₂CH₂COOH), 26.3 (2C, NCH₂CH₂CH₂CH₂N), 25.0 (24C, NCH₂CH₂CH₂N), 24.6 (4C, NCH₂CH₂CH₂N). IR (KBr) ν cm⁻¹ = 1572 (C=O, a); 1413 (C=O, s). ESI-MS m/z : 3993.6 [M + H⁺]. Anal. Calcd for C₁₈₄H₄₆₄N₃₀O₆₄ (MW 3992.82).

Detailed information on the synthesis and characterization of the other generations can be obtained as Supporting Information.

NMR Spectroscopy. ¹H NMR and ¹³C NMR spectra were recorded on a Bruker AM 400 spectrometer at 400.13 and 100.62 MHz respectively.

FT-IR Spectroscopy. IR samples were prepared according to the KBr technique and were measured on a Perkin-Elmer 1605 Series FT machine.

Electrospray Analysis. Electrospray mass spectra were recorded on a API 300 MS/MS mass spectrometer (PE-Sciex, Foster City, USA). Preparation and measurement of the samples were performed analogous to the ones in previous publications.²³ The resulting ESI-MS spectra of generations 1–4 are shown in Figure 2. The mass spectrometer was used

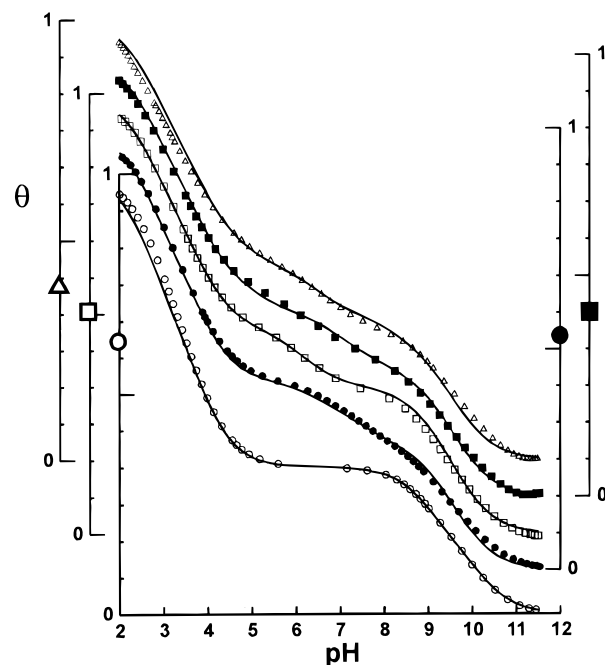


Figure 4. Degree of protonation θ as a function of pH at 1.0 M KCl for the carboxylate functionalized poly(propyleneimine) dendrimers. Generations 1 (○), 2 (●), 3 (□), 4 (■), and 5 (△) are shown with a shift along the degree of protonation; the corresponding axis is shown with the same symbol. Lines through the data points are fits based on the Ising model.

in positive ion mode. Electrospray data were deconvoluted to yield the resulting ESI-MS spectra. Up to generation 4 (almost) no defect structures are observed, indicating that the purity of the compounds is very high. The spectra indicate the pure character of the dendrimers with parent peaks that can be assigned to the correct theoretical mass, no structural defects are observed. ESI-MS analysis of generation 5 proved unsuccessful under the used conditions; however, this is not due to an ill-defined structure as can be derived from the NMR spectroscopy and FT-IR results.

Potentiometric Titrations. Potentiometric titrations were carried out at 22 ± 1 °C, with a VIT90 Video Titrator and a combined electrode (Radiometer, Copenhagen). Electrode calibration was based on buffer solutions pH 7 and 10 (Titrisol, Merck). The titrations were performed with HCl and KOH (Titrisol, Merck) in 0.1 and 1.0 M KCl (p.a., Merck). The condition of constant ionic strength throughout the measurement was fulfilled using acid and base in the same concentrations as the supporting electrolyte. The contribution of the dendrimer concentration to the ionic strength was negligible ($<10^{-2}$ M sites). More detailed information about both the method and the analysis is given in a previous publication.¹⁵ For all titrations the degree of protonation could be reproduced within an error of $\pm 1\%$ within the pH range 2.5–11.

The nitrogen content, necessary to calculate the degree of protonation, was determined with a TN 3000 nitrogen analyzer (Euroglas, Delft, The Netherlands).

For the first generation with only six sites, the protonation behavior has also been modeled with an expression involving protonation constants for the macroscopic protonation states.²⁴ The accuracy of the obtained macroconstants was 0.1 pH units. In a nonlinear least-squares fit procedure, the total concentration of sites was used as an additional fit variable and reproduced the total number of sites determined with the TN 3000 nitrogen analyzer to within 1%.

Titration Results and Discussion

The protonation behavior of the five generations of carboxylate-functionalized poly(propyleneimine) dendrimers in 1.0 M KCl is presented in Figure 4. The

Table 1. Parameters Used to Fit the Ising Model to the Protonation Curves of All Five Generations of Carboxylate-Functionalized Poly(propyleneimine) Dendrimers Together at One Ionic Strength^c

parameters	0.1 M KCl	1.0 M KCl
$pK^{(COO)}$	3.8 (4.3 ^a)	4.1
$pK^{(N,i)}$	9.2	9.5
$pK^{(N,o)}$	10.0 (10.1 ^a)	9.6 (9.9 ^a)
$pK^{(N1)}$	9.0	9.2
$pK^{(N2)}$	8.0	8.8
ϵ_1	0.6 ^b	0.6 ^b
ϵ_2	1.0 ^b	1.0 ^b
ϵ_{NCOO}	0.5	0.5
ϵ'	0.6	0.6

^a Results of fit method in which **1** was treated separately from all other generations. ^b Fixed values in fit method, based on previous work.^{15,25} ^c Extra $pK^{(COO)}$ and $pK^{(N,o)}$ values are used to fit the titration data of the first generation, presented in the table between brackets. In all cases the pair interactions between the amines with spacer of four and three carbon atoms (ϵ_1 and ϵ_2) are fixed at values obtained from previous work.^{15,25} Furthermore, at low ionic strength ϵ' is fixed at the value obtained at 1.0 M because of lack of data at 0.1 M

curves for low ionic strength (0.1 M KCl) are given in the Supporting Information. The titration curve for the highest generation at low ionic strength showed hysteresis at pH > 10, but for the other generations no problems were observed.

For the first generation, the rather pronounced difference in dissociation constant between carboxylic groups and amines (4 vs 9–10) yields two distinct steps in their titration curves. This results in low buffer capacity after protonation of the two inner amines, one-third of all sites. Only below pH 4 are the carboxylic groups involved in protonation. Since, for all generations, these end groups experience the same interactions with the interior amines, the last parts of the titration curves overlap.

The pairwise interactions between neighboring groups lead to onion-like shell protonation behavior, such as already observed for the amine-functionalized poly(propyleneimine) dendrimers.¹⁵ The outermost shell of tertiary amines protonates first; these amines appear to have a higher pK value due to the neighboring carboxyl groups.

The parameters, as presented in Figure 3, are used to fit the experimental curves for all generations at one ionic strength simultaneously (see Table 1). The results are shown as continuous lines through the data points in Figure 4. The quality of the fit is similar for data in 0.1 M KCl (see Supporting Information), but it seems that for low ionic strength additional pair interactions between nonneighboring groups need to be included. We have however no rational ideas about the size of these additional interactions and therefore leave them out.

Nearly all parameters were kept at the same value for all generations of dendrimers, except for $pK^{(N,o)}$ and $pK^{(COO)}$ for the first generation. The value obtained for $pK^{(COO)}$ at high ionic strength for the first generation was found to be the same as the value for the higher generations. The $pK^{(N2)}$ value found for the inner amines of the two higher generations is of the same value as $pK^{(3)}$ obtained earlier for the amine-functionalized poly(propyleneimine) dendrimers.¹⁵ The interior of the dendrimer does not seem to be affected by the presence of the carboxylic end groups. This indicates that the carboxylate end groups are not folding back, as that would have its effect on the interior.

Table 2. The Six Successive Macroscopic Protonation Constants pK_i at both 0.1 and 1.0 M KCl Presented for the First Generation Carboxylate-Functionalized Poly(propyleneimine) Dendrimer 1^c

<i>i</i>	pK_i			
	0.1 M		1.0 M	
	fit ^a	Ising ^b	fit ^a	Ising ^b
1	10.38	10.40	10.16	10.20
2	9.16	9.20	9.00	9.00
3	4.40	4.40	4.21	4.20
4	3.72	3.85	3.59	3.65
5	3.21	3.15	3.12	2.95
6	2.60	2.60	2.52	2.40

^a Obtained by direct fitting of experimental data. ^b Calculated from the Ising model parameters given in Table 1. ^c All fitted pK_i values are obtained with accuracy within ± 0.1 pH units as a result of direct fitting of experimental data. Also presented are the macroscopic protonation constants calculated from the Ising parameters given in Table 1

Also, the pair interactions ϵ_1 and ϵ_2 between the amine groups are the same as observed for other polyamines and oligoamines.^{25,26} It has been argued there that these interactions are entirely along the chain and hence do not vary with ionic strength. The interactions ϵ_{NCOO} between the amines and carboxylic groups also do not change with ionic strength; see Table 1. Titration data at low ionic strength are only available up to $\theta \approx 0.8$. This hampered the correct estimation of the parameter ϵ' , the pair interaction between neighboring carboxylic groups. Therefore, its value was fixed at the value obtained for 1.0 M.

The potentiometric titrations show different dependence on ionic strength for the first generation compared to the other generations. The behavior of the smallest dendrimer is in line with that of EDTA.²⁷ The first generation has been treated as a hexaprotic acid, and the resulting macroscopic pK values are presented in Table 2. The macroscopic pK values are all 0.1–0.2 lower for the higher ionic strength, the titration curve shows an overall shift toward lower pH. This is completely opposite to the observed protonation behavior for the amine-functionalized poly(propyleneimine) dendrimers.¹⁵ The macroscopic protonation constants are also calculated with the Ising model parameters given in Table 1. It shows that the Ising model is a good substitution for directly fitting of experimental data with macroscopic protonation constants. The latter approach is only applicable for small molecules as the number of fit parameters grows linearly with the number of sites. The Ising model however has the advantage that only a few parameters are needed to predict the protonation behavior of systems with a large number of sites. In the present article the protonation behavior of the highest generation (126 ionizable groups) is described with only nine parameters.

The titration curves of all other generations than the first generation, on the contrary, show a crossing point with changing ionic strength rather than a parallel shift along the pH axis. This is demonstrated in Figure 5 for the fourth generation, where the curves in both 0.1 and 1.0 M KCl are shown together with two additional ionic strengths, 0.2 and 0.5 M. This phenomenon is reflected in the fitted pK values. There one observes that those for most of the shells increase with ionic strength whereas the one, $pK^{(N,o)}$, for the outer amine shell decreases with ionic strength. At the crossing point the opposite changes in $pK^{(N,o)}$ on the one side, and $pK^{(N,i)}$,

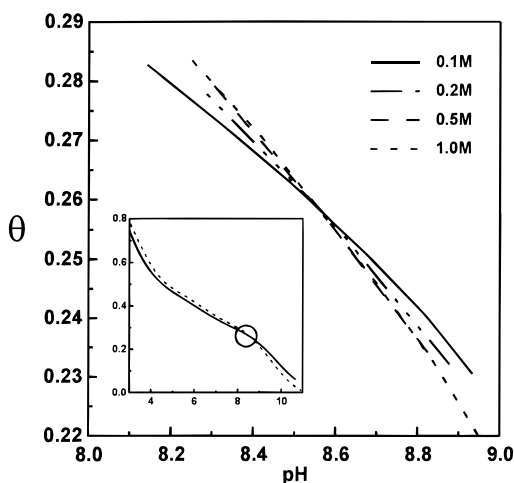


Figure 5. Influence of ionic strength on the protonation behavior shown over the whole pH range for the fourth generation in 0.1 and 1.0 M KCl and at two additional ionic strengths (0.2 and 0.5 M) around the crossing point. The trends are representative for all other higher generations than the first generation. They all feature a crossing point different from the point of zero charge at half protonation. Instead the crossing point is observed after protonation of the outermost shell of amine groups, theoretically at $\theta = n/2(2n - 2)$, where n is the number of (carboxylate) end groups.

$pK^{(N1)}$ and $pK^{(N2)}$ on the other side, compensate each other. The interaction parameters do not vary with ionic strength; see Table 1.

The opposite types of charges on these dendrimeric structures make it possible to create a negatively charged surface and a core with a high positive charge density. All generations exhibit amines and carboxylate groups in a $(n - 2):n$ ratio, where n is the number of carboxylate end groups ($n = 4, 8, 16, 32, 64$). The amine groups will be protonated at higher pH than the carboxylate groups. Therefore, at a degree of protonation $\theta \approx 0.5$, all amine groups are positively charged and all carboxylate groups are negatively charged. This is the so-called zero point of charge or the isoelectric point. For the higher generations this point is around pH 4.5 at 0.1 M, and 4.7 at 1.0 M. The titration curves for the different ionic strengths cross in a common intersection point. The crossing point, shown in detail in Figure 5 for four different ionic strengths, occurs at $\theta \approx 0.25$ for the fourth generation, which corresponds to full protonation of the second shell. For the second, third, and fifth generations the crossing point is also observed at a degree of protonation which corresponds to a fully protonated outermost shell of amines (theoretically at $\theta = n/2(2n - 2)$). This finding is remarkable, as this crossing point and the zero point of charge do not coincide. In many other systems this common crossing point coincides with the point of zero charge, such as, for example, for proteins²⁸ and metal oxides.²⁹

In summary, an alternative and highly efficient synthesis route for carboxylate-functionalized poly(propyleneimine) dendrimers is described, which does not have the drawbacks (structure defects, hygroscopic, high salt concentration) of other routes. The protonation behavior of these potential complexing agents is studied with potentiometric titrations in 0.1 and 1.0 M KCl solutions. The dendrimers show the characteristic on-ionlike shell protonation behavior. The titration curves for different ionic strengths cross at pH 8.5, which is exactly after protonation of the outermost shell of

amines ($\theta \sim 0.25$). This crossing point is found not to be identical to the point of zero charge $\theta \sim 0.5$, which is observed at pH 4.5. The Ising model was used to rationalize the complete set of titration data simultaneously for five generations with only one microscopic pK value for each additional shell and a set of three nearest neighbor pair interaction parameters.

Acknowledgment. The authors would like to thank Joost van Dongen (Eindhoven University of Technology) for performing the ESI-MS measurements and Euroglas for the analysis of the nitrogen contents. DSM Research is acknowledged for providing the poly(propyleneimine) dendrimers and for an unrestricted research grant.

Supporting Information Available: Text giving syntheses of **1–3** and **5** and a figure showing the degree of protonation as a function of pH for **1–5** at 0.1 M. This material is available free of charge via the Internet at <http://pubs.acs.org>.

References and Notes

- (1) Tomalia, D. A.; Naylor, A. M.; Goddard, W. A., III. *Angew. Chem., Int. Ed. Engl.* **1990**, *29*, 138.
- (2) Frechet, J. M. J. *Science* **1994**, *263*, 1710.
- (3) Newkome, G. R.; Moorefield, C. N.; Vogtle, F. *Dendritic Molecules: Concepts, Syntheses, Perspectives*, VCH: New York, 1996, and references therein. Ardoin, N.; Arstruc, D. *Bull. Soc. Chim. Fr.* **1995**, *132*, 875. Zimmermann, S. C.; Zeng, F. Z. *Chem. Rev.* **1997**, *97*, 1681.
- (4) Bosman, A. W.; Janssen, H. M.; Meijer, E. W. *Chem. Rev.* **1999**, *99*, 1665.
- (5) Muijselaar, P. G. H. M.; Claessens, H. A.; Cramers, C. A.; Jansen, J. F. G. A.; Meijer, E. W.; De Brabander-van den Berg, E. M. M.; Van der Wal, S. J. *High Resolut. Chromatogr.* **1995**, *18*, 121.
- (6) De Brabander-van den Berg, E. M. M.; Meijer, E. W. *Angew. Chem., Int. Ed. Engl.* **1993**, *32*, 1308.
- (7) The nomenclature of the poly(propyleneimine) dendrimers is the following: DAB-dendr-(NH₂)_n; DAB stands for the core molecule 1,4-diaminobutane and *dendr* for the poly(propyleneimine) dendrimer, whereas (NH₂)_n stands for amine end groups and n is the number of end groups. Upon modification, e.g. DAB-dendr-(COO⁻Li⁺)_n is the notation for the carboxylate-functionalized poly(propyleneimine) dendrimers with Li⁺ as the counterion. Again n is used to represent the number of end group functionalities.
- (8) Bosman, A. W.; Schenning, A. P. H. J.; Janssen, R. A. J.; Meijer, E. W. *Chem. Ber./Recl.* **1997**, *130*, 725.
- (9) Marcus, R. A. *J. Phys. Chem.* **1954**, *58*, 621.
- (10) Katchalsky, A.; Mazur, J.; Spitnik, P. *J. Polym. Sci.* **1957**, *23*, 513.
- (11) Kitano, T.; Kawaguchi, S.; Ito, K.; Minakata, A. *Macromolecules* **1987**, *20*, 1598.
- (12) Kawaguchi, S.; Kitano, T.; Ito, K.; Minakata, A. *Macromolecules* **1990**, *23*, 731.
- (13) Smits, R.; Koper, G. J. M.; Mandel, M. *J. Phys. Chem.* **1993**, *97*, 5745.
- (14) de Groot, J.; Koper, G. J. M.; Borkovec, M.; de Bleijser, J. *Macromolecules* **1998**, *31*, 4182.
- (15) van Duijvenbode, R. C.; Borkovec, M.; Koper, G. J. M. *Polymer* **1998**, *39*, 2657.
- (16) Koper, G. J. M.; Genderen, M. H. P. van; Elissen-Roman, C.; Baars, M. W. P. L.; Meijer, E. W.; Borkovec, M. *J. Am. Chem. Soc.* **1997**, *119*, 28, 6512.
- (17) Harris, F. E.; Rice, S. A. *J. Phys. Chem.* **1954**, *58*, 725.
- (18) Borkovec, M.; Koper, G. J. M. *Macromolecules* **1997**, *30*, 2151.
- (19) Tanford, C.; Kirkwood, J. C. *J. Am. Chem. Soc.* **1957**, *79*, 5333.
- (20) Honig, B.; Nicholls, A. *Science* **1995**, *268*, 1144.
- (21) Bashford, D.; Karplus, M. *J. Phys. Chem.* **1991**, *95*, 9556.
- (22) Borkovec, M.; Koper, G. J. M. *Prog. Colloid. Polym.* **1998**, *109*, 142.
- (23) Hummelen, J. C.; Dongen, J. L. J. van; Meijer, E. W. *Chem.-Eur. J.* **1997**, *3*, 1489.

- (24) Borkovec, M.; Koper, G. J. M. *Ber. Bunsen-Ges. Phys. Chem.* **1996**, *100*, 764.
- (25) Borkovec, M.; Koper, G. J. M. *J. Phys. Chem.* **1994**, *98*, 6038.
- (26) Private communications.
- (27) Smith, R. M.; Martell, A. E. *Critical Stability Constants*; Plenum Press: New York, 1989.
- (28) Tanford, C.; Hauenstein, J. D. *J. Am. Chem. Soc.* **1956**, *78*, 5287.
- (29) Schudel, M.; Behrens, H.; Holthoff, H.; Kretzschmar, R.; Borkovec, M. *J. Colloid Interface Sci.* **1997**, *196*, 241.

MA990968J