

Charge Determination of Cationic Polyelectrolytes by Visual Titrimetry and Spectrophotometry

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Polyelectrolyte titration, which was called colloid titration is based on the stoichiometric reaction between oppositely charged polyelectrolytes. This can be used, for instance, to determine the charge density of a cationic polyelectrolyte, using an anionic polyelectrolyte of known charge density, such as potassium polyvinyl sulfate (PPVS). The technique requires a suitable method of end-point detection and there are several possibilities. In this work, two methods have been investigated: visual titrimetry based on the color change of a cationic dye (o-toluidine blue, o-Tb) and spectrophotometry based on the absorbance change corresponding to the color change of the same dye. These have been applied to several cationic polyelectrolytes with different charge density and molecular weight. In all cases, the cationic charge was due to quaternary nitrogen groups. In the case of cationic dye, it was shown that the sharpness depends on the charge density of cationic polyelectrolyte. With the polyelectrolytes of lower charge density, the binding to PPVS is weaker and binding of the dye to PPVS can occur before all of the polyelectrolyte charge has been neutralized. However, by carrying out titrations at several polyelectrolyte concentrations, good linear relationships were found, from which reliable charge density values could be derived. Effects of pH and ionic strength were also briefly investigated. For cationic polyelectrolytes (copolymers of acrylamide and dimethylaminoethyl acrylate), there was some loss of charge at higher pH values, probably as a result of hydrolysis. Increasing ionic strength causes a less distinct color change of o-Tb, as a result of weaker electrostatic interactions.

Key Words : Charge density, Cationic polyelectrolytes, Cationic dye, Potassium Polyvinyl sulfate, Visual titrimetry, Spectrophotometry

1. Introduction

Naturally occurring and synthesized polyelectrolytes have been extensively applied in a wide variety of industrial fields, e.g., as polymeric flocculants in water and effluent treatment, papermaking and many other examples, as aids in the secondary recovery of petroleum and to reduce turbulent friction of water, and as components in foods, cosmetics and drugs^{1,2)}. In the application as polymeric flocculants, their charge density is often of great importance³⁻⁵⁾. It is frequently found that the optimum dosage of the flocculant is close to that required to neutralize the surface charge carried by the particles⁶⁾, although there are many im-

portant exceptions⁷⁾. Most particles and dissolved organic materials (DOM) in aqueous system are anionic in character and can be effectively removed by cationic additives, such as hydrolyzing metal salts and cationic polyelectrolytes. In these cases there is a stoichiometric relationship between the anionic charge carried by the particles or DOM and the cationic charge of the additive^{8,9)}, therefore, optimum dosages can be predicted if the appropriate charge information is available⁹⁾. For these reasons, the charge density of natural and synthetic polyelectrolytes is of interest and convenient experimental methods are needed.

Polyelectrolyte titration, which was originally called colloid titration and introduced by Terrayama¹⁰⁾, is a useful method for the determination of charge carried by dissolved macromolecules²⁾. This method is based on the stoichiometric reaction between oppositely

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charged polyelectrolytes. For the end point detection, Terrayama used the interaction of the cationic indicator, o-toluidine blue (o-Tb, 3-amino-7-dimethylphenothiazin-5-ium) with anionic polyelectrolyte poly (potassium vinyl sulfate) (PPVS), which leads to a metachromatic color change of o-Tb from blue to red violet. It has been published that this visual titrimetry often depends on the person performing the titration because the color change is not very sharp, and the reproducibility is quite poor for the polyelectrolytes of low charge density²⁾. However, it is considered that if the absorbance change (i.e., spectrophotometry¹¹⁾) corresponding to a color change of o-Tb with the titration for the end point detection is utilized, more accurate and reproducible charge densities of polyelectrolytes can be determined. Alternatives to these methods include potentiometry¹²⁾, conductimetry¹³⁾, phototitrimetry¹⁴⁾, streaming current detector (SCD) method^{9,15)}. Where the reaction between oppositely charged polyelectrolytes leads to the formation of a precipitate, end point detection by particle detection method¹⁶⁾ can also be used.

In the present paper, the charge densities of several cationic polyelectrolytes of different charge density and molecular weight have been determined using visual titrimetry and spectrophotometry as the method of end point detection and the results between both methods have been compared.

2. Materials and Methods

2.1. Reagents and apparatus

Analar grade reagents were used and the solutions were prepared with deionized water from an Elga 'Spectrum' unit.

Cationic polyelectrolytes used were commercial products, kindly supplied by Allied Colloids Ltd. (Now Ciba Speciality Chemicals), UK. These were copolymers of acrylamide and dimethylaminoethyl acrylate (DMAEA) (the latter being quaternized with methyl chloride) (P-A ~ P-G), poly(2-hydroxypropyl-N,N-dimethylammonium chloride) (P-H) and poly(diallyldimethylammonium chloride) (P-I and P-J). P-A ~ P-G were supplied as solids and prepared as 0.1% aqueous solutions by wetting 0.1 g of the solid with 2 mL of methanol, adding 98 mL of water and agitating for about 12 hrs. P-H (a 50% active solution), and P-I and J (a 40% active solution) were also prepared as 0.1% aqueous solutions by diluting the

products supplied with water. All of the 0.1% solutions were used within one week of preparation. The properties of the polyelectrolytes used are given in Table 1.

PPVS was obtained from Aldrich Chemical Co., UK. The solution was prepared by dissolving 180 mg in 1 L of water. The charge density of this solution was standardized with a cationic surfactant, cetyltrimethylammonium bromide (CTAB), obtained from BDH Co., UK and prepared as a 1 meq/L solution.

Ortho-toluidine blue (o-Tb) was obtained from BDH Co., UK and prepared as a 0.01% aqueous solution.

Buffer solutions (1 mmol/L) were used in investigations of pH effects. These were acetic acid/sodium acetate for pH 5, NaH₂PO₄/Na₂HPO₄ for 7, and NH₄OH/NH₄Cl for pH 9.

A UV/Vis spectrophotometer (Camspec M 350, UK) was used for all absorbance measurements and absorption spectra. Continuous titration of samples in a 10 mm quartz cuvette was carried, using a syringe pump (Cole-Palmer Instrument Co., USA) and a magnetic cuvette stirrer (Rank Brothers Ltd., UK). A Cranwell Universal pH 950 pH meter was used for pH measurements.

2.2. Procedure

Visual titrimetry: The PPVS solution was standardized with CTAB. A beaker containing 30 mL of water, 3 mL of CTAB (1 meq/L), 100 μ L of o-Tb and a magnetic stirrer bar, was placed on a magnetic stirrer. The solution was titrated with PPVS until the

Table 1. Properties of cationic polyelectrolytes used

Polyelectrolyte % Cationic	Intrinsic Viscosity (dL/g)	Molecular Weight ^a
P-A	20	6.5
P-B	25	12
P-C	40	9
P-D	40	12
P-E	40	14
P-F	80	8
P-G	80	12
P-H	100	0.2
P-I	100	0.2
P-J	100	1.0

^acalculated from intrinsic viscosity values given by the manufacturer, using the Mark-Houwink equation ($\eta = KM^a$), with estimates of the constants K (= 7.3 $\times 10^{-4}$ dL/g) and a (= 0.66) taken from Gill and Herrington¹⁷⁾.

color of the solution changed from blue to red violet. The titration was repeated with a blank sample, containing the same amount of indicator. From the difference in the volumes needed, the PPVS concentration could be calculated. This is necessary due to the fact commercial PPVS is rather impure and leaves some solid residue when dissolved in water.

The titration procedure was repeated for solution of cationic polyelectrolytes A-J (usually 1 mL of 0.1% solution), using PPVS of pre-determined charge density according to the above procedure. From the results, the charge densities of the polyelectrolytes (meq/g) could be calculated.

Spectrophotometry: Between 1.65-1.8 mL of water or buffer, 0.2 mL of o-Tb solution, 0-0.15 mL of polyelectrolyte and magnetic stirrer bar were placed in a 10 mm quartz cuvette, which was inserted into the spectrophotometer on the cuvette stirrer unit (the sample was then in the light beam of the spectrophotometer). The cuvette contents were stirred and PPVS solution (0.914 meq/L) was introduced from the syringe pump at a flow rate of 0.06 mL/min. The spectrophotometer output was connected to a computer, so that the absorbance at 635 nm could be monitored continuously. The syringe pump and data output (computer) were started simultaneously and stopped when the graph showed little further change in absorbance. The effect of ionic strength on the determination of charge density of cationic polyelectrolyte was carried out with polyelectrolyte P-J, over a range of NaCl concentration (0-200 mM). The experimental set-up is

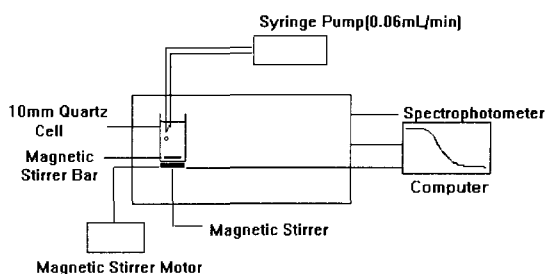


Fig. 1. Schematic diagram of experimental set-up by spectrophotometry.

shown in Fig. 1.

3. Results and Discussion

3.1. Visual titrimetry

In order to determine the charge density of cationic polyelectrolytes by titration with the anionic polyelectrolyte PPVS, the charge density of PPVS should be determined. It was standardized against a CTAB solution (1 meq/L) using o-Tb as a visual indicator. CTAB, which is a long chain surfactant, binds strongly to high surface charge anionic polyelectrolyte, because of hydrophobic attraction between CTA⁺ chains, after their charges have been neutralized by association with anionic sites on the polyelectrolyte chain¹². For this reason, CTAB can be used to standardize PPVS solutions.

The values of charge densities for polyelectrolytes against PPVS by visual titrimetry are summarized in Table 2. The polyelectrolytes were dissolved in de-

Table 2. The charge densities of cationic polyelectrolytes in water (at about pH 5)

Polyelectrolyte	Charge density (meq/g)		
	Visual titrimetry	Spectrophotometry	
		Upper break point (r^*)	Lower break point (r^*)
P-A	0.863	0.824 (0.998)	0.832 (0.997)
P-B	0.958	0.928 (0.999)	0.936 (0.997)
P-C	1.805	1.733 (0.997)	1.759 (0.998)
P-D	1.657	1.568 (0.999)	1.585 (0.998)
P-E	1.561	1.521 (0.999)	1.538 (0.997)
P-F	3.689	3.312 (0.998)	3.419 (0.999)
P-G	3.268	3.014 (0.999)	3.163 (0.998)
P-H	7.058	6.635 (0.999)	6.832 (0.999)
P-I	6.479	6.173 (0.998)	6.245 (0.999)
P-J	5.271	4.932 (0.999)	5.036 (0.998)

* Correlation coefficient.

ionized water, giving a pH value of about 5 in all cases. The results are in line with the order of values given in Table 1. However, it can be seen that the values are a little different for the polyelectrolytes of the same charge density but different molecular weight (e.g., P-C~P-E, P-F and P-G, P-I and P-J), showing a little decrease with increasing molecular weight. The reason is not evident, but it is considered to be that the exact amounts of raw materials required for the corresponding cationic charge and molecular weight are not added.

3.2. Spectrophotometry

3.2.1. Charge densities of cationic polyelectrolytes in water

The binding of o-Tb to PPVS produces a distinct color shift from blue (absorption maximum at 635 nm) to red-violet (530 nm)¹¹⁾. In this work, the reduction in the absorbance at 635 nm was followed, because the change in the absorbance at 635 nm is more sensitive than that at 530 nm.

Titrations at different concentrations (0-75 mg/L) were carried out in order to determine charge densities of cationic polyelectrolytes in water. Spectrophotometric titration curves of P-B, P-D and P-G of different charge density but same molecular weight measured in water by procedure are shown in Fig. 2(a-c). The change in the absorbance of the indicator is plotted against the volume of PPVS. The curves are progressively displaced to the left with increasing concentration of polyelectrolyte in the cell. The curve for 0 mg/L is the blank and shows the amount of PPVS required to bind to the indicator.

As shown in Fig. 2(a), for P-B of low charge density (20% cationic, <1 meq/g in visual titrimetry), the titration curves show a broad region, where the decrease in the absorbance of o-Tb is rather gradual and no distinct equivalence point is apparent. During the titration of polyelectrolyte B against PPVS, o-Tb is binding to the titrant in competition with polyelectrolyte B, owing to a rather low binding affinity of the latter to PPVS. This is consistent with large mismatch of spacing between charge centers of polyelectrolyte B (low charge density) and PPVS^{18,19)}, which has a much charge density. This effect leads to some uncertainty in locating the equivalence point, compared to, say, the SCD method^{9,15)}, in which the indicator is not used.

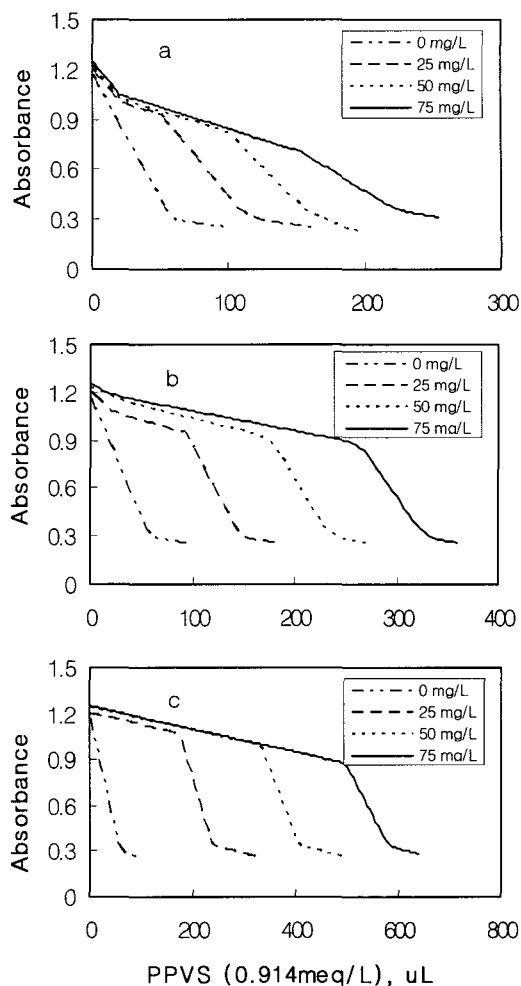


Fig. 2. Spectrophotometric titration curves of cationic polyelectrolytes P-B(a), P-D(b) and P-G(c), showing absorbance at 635 nm versus amount of PPVS solution added. Different curves correspond to different polyelectrolyte concentrations.

By contrast, P-G of high charge density (80% cationic, >3 meq/g in visual titrimetry), shows titration curves with sharp points (Fig. 2(c)) and the absorbance of o-Tb changes very little during the titration, until the end-point is reached. The slow decrease in the absorbance, before the sharp break-point, is simply due to the dilution which occurs during the titration. In this case, it can be concluded that the titration proceeds according to the proposed mechanism (i.e., the anionic polyelectrolyte-dye binding only takes place after the polyelectrolyte complex formation is virtually complete) and that the necessary condition for the ra-

tio of the two equilibrium constants, $K_1 \gg K_2$ is fulfilled¹⁴⁾ (K_1 , the equilibrium constant for the reaction between oppositely charged polyelectrolytes and K_2 , the equilibrium constant for the reaction between anionic polyelectrolyte and cationic dye). Fig. 2(b) shows the titration curves for P-D, of intermediate charge density (40% cationic, 1.5-2.0 meq/g in visual titrimetry), and it is clear that the shape of the titration curves lies between those of P-B and P-G.

The titration curves for 50 mg/L solutions of polyelectrolytes with same charge density but different molecular weight given in Table 1 are shown in Fig. 3(a-c). As shown in Fig. 3(a-c), these curves show a

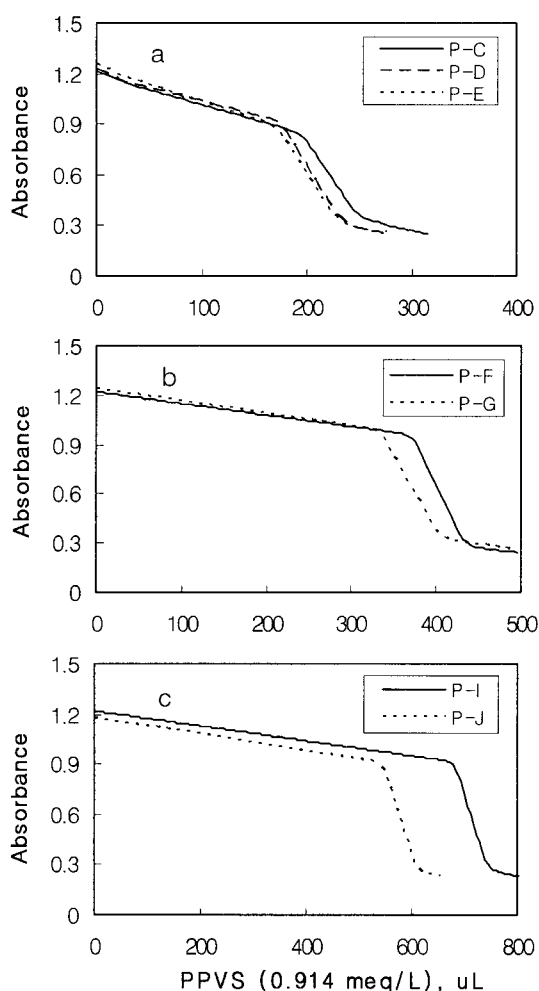


Fig. 3. Spectrophotometric titration curves for 50 mg/L solutions of polyelectrolytes with same charge density but different molecular weight given in Table 1 (a: 40% cationic, b: 80% cationic, c: 100% cationic).

similar shape in each group, indicating that molecular weight has little influence on the binding of the polyelectrolytes to PPVS, although the equivalence point (i.e., the charge densities of polyelectrolytes in each group) is a little different, like as visual titrimetry.

Two end points can be derived from these curves. This is illustrated by the titration curve for 50 mg/L of P-D in Fig. 4. The upper (1) and lower (2) break points can be determined by the intersection of the drawn lines as shown. By plotting the upper (1) and lower (2) break point against titrant volume, linear plots are obtained, with correlation coefficients very close to 1. The plots for P-B, P-D and P-G are shown in Fig. 5. The slope of these lines can be used to derive the charge densities of the polyelectrolytes

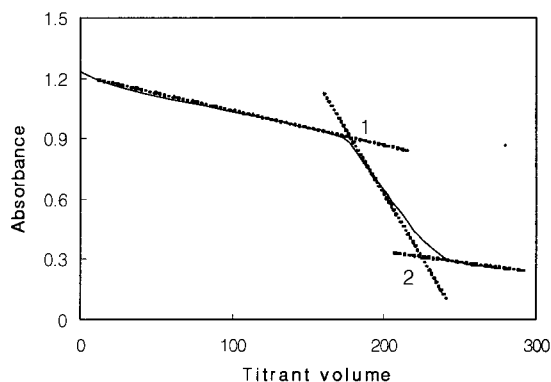


Fig. 4. Illustration of upper (1) and lower (2) break points for a polyelectrolyte titration curve.

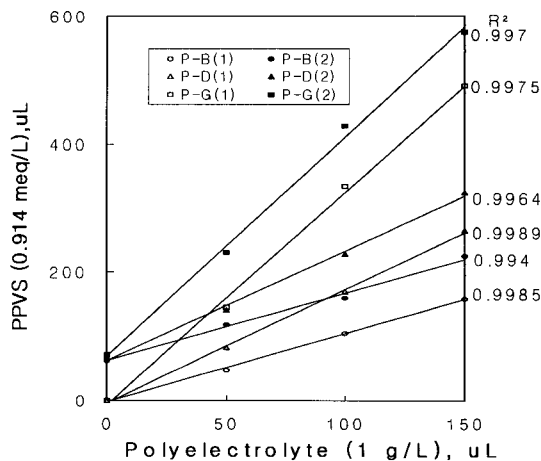


Fig. 5. Plots of end points (1 and 2) determined as in Fig. 4 for P-B, P-D and P-G at different concentrations. R^2 values for linear regression are also shown.

in more reliable manner than by simply using the break point value for one polyelectrolyte concentration.

The charge densities of cationic polyelectrolytes in water (about pH 5) obtained in this way are summarized in Table 2, together with the correlation coefficients. As shown in Table 2, the results from the upper and lower break points give similar values and the high correlation coefficients show that in all cases there is a very linear relationship, even when the reaction of o-Tb and PPVS occurs before the break points. The charge densities obtained for cationic polyelectrolytes are in line with the order of those given by the manufacturer, but are a little lower than those by visual titrimetry.

3.2.2. Effect of pH

Table 3 shows the results by spectrophotometry for the case where the polyelectrolytes were in buffer solutions (1 mM) at pH 7. It can be seen the charge densities for P-A ~ P-G determined at pH 7 are lower than those in water (about pH 5). This is not expected for quaternized materials, which should retain their charge density up much higher pH values. However, it is known that copolymers, such as P-A ~ P-G are subject to hydrolysis and loss of cationic group and that this tendency increases with increasing pH²⁰. In contrast to this, the charge densities for P-H ~ P-J at pH 7 are similar to those in water as expected.

Table 3. Charge densities of cationic polyelectrolytes in buffer solutions at pH 7 determined by spectrophotometry

Polyelectrolyte	Charge density (meq/g)	
	Upper break point (r)	Lower break point (r)
P-A	0.785 (0.997)	0.793 (0.997)
P-B	0.823 (0.997)	0.833 (0.998)
P-C	1.453 (0.999)	1.462 (0.999)
P-D	1.407 (0.999)	1.412 (0.998)
P-E	1.270 (0.997)	1.268 (0.998)
P-F	3.015 (0.998)	3.116 (0.999)
P-G	2.745 (0.999)	2.756 (0.998)
P-H	6.655 (0.999)	6.649 (0.999)
P-I	6.087 (0.999)	6.094 (0.999)
P-J	4.885 (0.999)	4.896 (0.999)

For P-G and P-I, the effect of pH on their charge densities were examined at different pH values (5, 7 and 9) (Fig. 6). The charge densities were calculated taking the upper break point as the end point (Table 4). For P-G, the charge density decreases with increasing pH. Again, this is not expected for a cationic polyelectrolyte with quaternary nitrogen groups, but may be a result of some hydrolysis of the cationic DMAEA units of the copolymer. In contrast to this, the measured charge densities of P-I in the pH range investigated are virtually independent of the pH as expected, since P-I is a salt of a strong base and is not

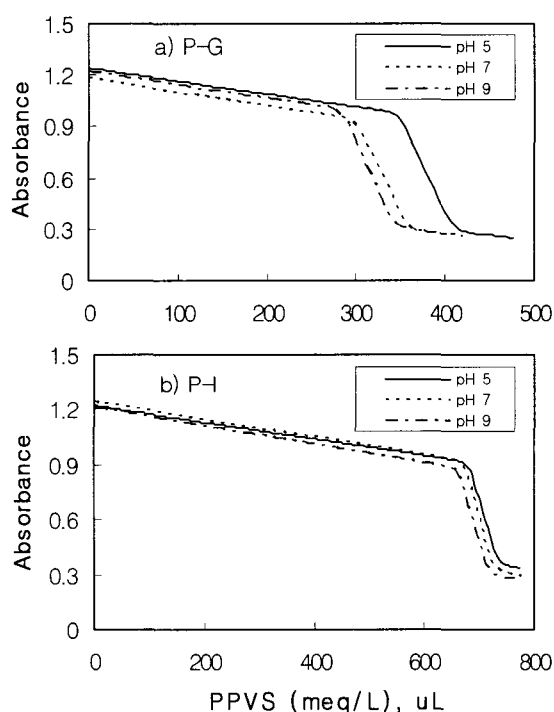


Fig. 6. Spectrophotometric titration curves for 50 mg/L solutions of P-G and P-I at different pH values (5, 7 and 9).

Table 4. The charge densities of P-G and P-I at different pH values (5, 7 and 9) determined by spectrophotometry

pH	Charge density (meq/g)	
	P-G	P-I
5	3.048	6.141
7	2.745	6.087
9	2.571	6.076

subject to hydrolytic loss of charge¹⁴⁾.

3.2.3. Effect of ionic strength

In the presence of higher concentrations of dissolved salts, i.e., at higher ionic strengths of the solution, the screening of electrostatic interactions is expected and this should influence polyelectrolyte titration procedures. The effect of ionic strength on the charge density of cationic polyelectrolyte was carried out with P-J, over a range of NaCl concentrations (0-200 mM) and the results are shown in Fig. 7. At higher salt concentrations, the titration curves are flatter in the region of end point and have a less pronounced break point. The absorbance change, even after the break point is quite small at 100 mM NaCl and greater, suggesting that the binding between o-Tb and PPVS is considerably weakened at these high salt concentrations, by screening effects. As a result, the equilibrium constant and thus the slope of the titration curves decreases. However, this flattening is only significant in the o-Tb/PPVS system for concentrations of monovalent salts than about 50 mM. Measurements are possible up to about 75 mM salt. The equivalence volume at break-point at low salt concentrations, is independent of the ionic strength. It is likely that divalent cations, such as Ca^{2+} , would have much greater effects and this can be a serious complicating factor in conventional polyelectrolyte titrations¹⁴⁾.

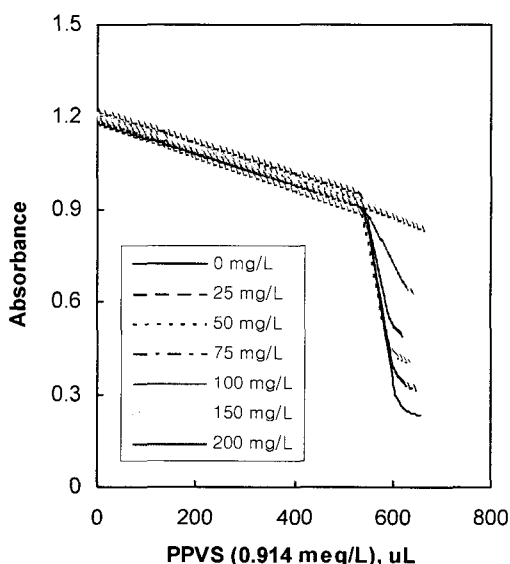


Fig. 7. Spectrophotometric titration curves for P-J (50 mg/L) at different concentrations of NaCl.

4. Conclusions

In order to determine the charge densities of several cationic polyelectrolytes of different molecular weight and charge density have been determined using visual titrimetry and spectrophotometry as the method of end point detection and the results between both methods have been compared.

The charge densities for each of cationic polyelectrolytes obtained by visual and spectrophotometry are in line with the order of those given by the manufacturer, but the values by the former are a little higher than those by the latter. The results obtained by visual titrimetry are less reliable, compared to spectrophotometry, especially for polyelectrolytes of low charge density, where the indicator binds to the titrant well before the equivalence point. The same problem affects the spectrophotometry, but good results can still be obtained by plotting data from a series of titrations with different concentrations of polyelectrolyte sample. With the spectrophotometry, the titration curves for polyelectrolytes of high charge density have a sharp break point, which becomes rather less sharp for polyelectrolytes of lower charge density. Nevertheless, reliable values of charge density can be obtained by this method. The charge densities obtained from upper and lower break points by spectrophotometry are almost identical and linear relationships hold, with high correlation coefficients (close to $r = 0.999$ for most polyelectrolytes) over the investigated concentration range.

The charge densities for polyelectrolytes P-A ~ P-G, which are copolymers of polyacrylamide and DMAEA, show some pH dependence, becoming lower as pH increases. This is not expected for cationic polyelectrolytes with quaternized groups, but the loss of charge may be due to hydrolysis. For polyelectrolytes P-H ~ P-J, the charge densities are virtually independent of pH in the range investigated, which is to be expected for a salt of a strong base.

The polyelectrolyte titration curves are flatter in the region of end point and have a less pronounced break point at higher salt concentrations, suggesting that the binding between o-Tb and PPVS is considerably weakened at high salt concentrations, by screening effects. As a result, the equilibrium constant and thus the slope of the titration curves decreases. However, the equivalence volume at break-point at low salt con-

centrations, is independent of the ionic strength.

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References

- 1) Moudgil, B. M. and R. Somasundaran, 1986, Flocculation, Sedimentation and Consolidation, Engineering Foundation, New York.
- 2) Tanaka, H. and Y. Sakamoto, 1993, Polyelectrolyte titration using fluorescent indicator. 1. Direct titration of anionic and cationic polyelectrolytes with 10^{-4} N standard solutions, *J. Polym. Sci.: Part A*, 31, 2687-2691.
- 3) Lee, S. Y. and J. Gregory, 1990, The effect of charge density and molecular mass of cationic polymers on flocculation kinetics in aqueous solution, *Water Supply*, 8, 11-17.
- 4) Kam, S. K., D. K. Kim, C. S. Moon, B. C. Ko and M. G. Lee, 2002, Flocculation characteristics of kaoline suspensions in water by cationic polyelectrolytes, *J. Environ. Sci.*, 11(2), 93-102.
- 5) Kam, S. K., D. K. Kim and M. G. Lee, 2003, Effects of polyelectrolyte dosage, kaoline particles and pH on flocculation of humic acid by cationic polyelectrolytes, *J. Environ. Sci.*, 12(8), 861-870.
- 6) Stumm, W. and C. R. O'Melia, 1968, Stoichiometry of coagulation, *J. Am. Water Works Assoc.*, 60, 514-539.
- 7) Gregory, J., 1996, Polymer adsorption and flocculation, *In* Finch, C. A. (ed.), *Industrial Water Soluble Polymers*, Royal Society of Chemistry, Cambridge, UK, pp. 62-75.
- 8) Tenney, M. W. and W. Stumm, 1965, Chemical flocculation of microorganisms in biological waste treatment, *J. Water Pollut. Control Fed.*, 37, 1370-1388.
- 9) Bernhardt, H. and H. Schell, 1993, Control of flocculants by use of a streaming current detector, *J. Water SRT-Aqua*, 42, 239-251.
- 10) Terrayama, H., 1952, Method of colloid titration (a new titration between polymer ions), *J. Polym. Sci.*, 8, 243-253.
- 11) Kam, S. K. and E. I. Cho, 1998, Charge determination of humic acid and humic extract by spectrophotometry, *Environ. Sci. (Bull. of the Korean Environ. Sci. Soc.)*, 2(2), 139-148.
- 12) Sequaris, J. M. and P. Kalabokas, 1993, Application of an iodide ion-selective electrode to the determination of anionic polyelectrolytes and colloids with a cationic surfactant, *Anal Chim Acta*, 281, 341-346.
- 13) Toei, K. and T. Kohara, 1976, A conductometric method for colloid titrations, *Anal. Chim Acta*, 83, 59-65.
- 14) Wassmer, K. H., U. Schroeder and D. Horn, 1991, Characterization and detection of polyanions by direct polyelectrolyte titration, *Makromol. Chem.*, 192, 553-565.
- 15) Kam, S. K., L. S. An and M. G. Lee, 1997, Comparison of flocculation- spectrophotometry and streaming current detector method to the control of flocculants for the removal of humic acid, *Environ. Sci. (Bull. of the Korean Environ. Sci. Soc.)*, 1(2), 137-144.
- 16) Gregory, J. and D. W. Nelson, 1984, A new optical method for flocculation monitoring, *In* Gregory, J. (ed.), *Solid-Liquid Separation*, Ellis Horwood, Chichester, pp. 172-182.
- 17) Gill, R. I. S. and T. M. Herrington, 1987, The flocculation of kaoline suspensions with cationic polyacrylamides of varying molar mass but the same cationic character, *Colloids & Surfaces*, 22, 51-61.
- 18) Gummow, B. D. and G. A. Roberts, 1985, Studies on chitosan-induced metachromasy, 2: the reversal of metachromasy by co-solutes, *Makromol. Chem.*, 186, 1245-1253.
- 19) Ortona, O., V. Vitagliano, R. Sartorio and L. Costantino, 1984, Spectrophotometric study of the interaction of poly(styrenesulfonic acid) with a metachromatic dye in methanol, *J. Phys. Chem.*, 88, 3244-3248.
- 20) Toussaint, A. F., 1987, The flocculation of calcium carbonate and its deposition onto cellulose fibres, PhD Thesis, University of London.