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Complexation between polyallylammonium cation and polystyrenesulfonate anion; the effect of ionic strength and electrolyte type

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Complexation between polyallylammonium cation and polystyrenesulfonate anion was investigated in aqueous solutions of binary 1:1 sodium electrolytes (NaX, X = F, Cl, Br, I, NO_3, ClO_4) by means of microcalorimetry, dynamic light scattering, electrokinetics and spectrophotometry. At lower molar ratios of monomer units charged polyelectrolyte complexes were formed. At molar ratios close to equivalence and at lower salt concentrations ($c(NAX)$ / mol dm^{-3} ≤ 0.1) flocculation occurred. The obtained precipitates contained approximately equimolar amounts of oppositely charged monomer units. At $c(NAX)$ / mol dm^{-3} ≥ 0.5 (X = NO_3, ClO_4) and in the case when the polycation was present in excess, the amount of positively charged monomer units in the precipitate was higher than that of negatively charged monomers (asymmetric neutralisation). In addition, the aggregation of positively charged complexes in concentrated solutions of all investigated electrolytes was noticed. The onset of aggregation was strongly anion specific. However, the aggregation of negatively charged complexes did not occur even at $c(NaX) = 3$ mol dm^{-3}. The composition of the insoluble products at equimolar ratio of monomer units and higher concentrations of $NaNO_3$ and $NaClO_4$ was dependent on the order of addition, indicating non-equilibrium interpolyelectrolyte neutralisation at all ionic conditions. At 25 °C and $c(NaClO_4) = 1$ mol dm^{-3} equilibrium was not reached after two months. By contrast, the supernatants showed no traces of free polyanion chains after being heated for a week at 60 °C. The pairing of monomer units was predominantly entropically driven, irrespectively of the type of reaction products formed (polyelectrolyte complexes, precipitates), and electrolyte type. The results obtained indicate that the

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overcharging is not an enthalpically demanding process. The calorimetric measurements also suggest that strong influence of counteranions on the composition of reaction product must be related with differences in ion distribution around polycation. However, despite rather similar energetics for complex and precipitate formation in the presence of various sodium salts a clear correlation of formation enthalpies with corresponding anion hydration enthalpies (Hofmeister series) was observed. Somewhat surprisingly, the titration calorimetry experiments have also revealed that the increase in electrolyte concentration affects the enthalpy of interpolyelectrolyte neutralisation negligibly.

1. Introduction

The first study of the reaction involving high-charge density linear synthetic polyelectrolytes (poly(4-vinyl-butylpyridonium) bromide and sodium poly(acrylate)) was reported by Fuoss and Sadek in 1949.[1] They noticed that the addition of one reactant into a solution of the other led to instant formation of colloid suspension, which at low molar ratio of oppositely charged monomer units ($r < 1$) showed no tendency of phase separation. At an approximately equimolar ratio of added monomers phase separation (flocculation) occurred and the addition of titrant above $r = 1$ did not affect the particle size. By inspecting the Br^{-} content in the precipitates, obtained by titration of polycation with polyanion and *vice versa*, the authors concluded that a metastable product containing high content of paired monomers had been formed. Fuoss and Sadek proposed that the process was driven by the entropically favourable expulsion of counterions from the polyion domain. They also suggested that the colloid particles bear an excess of titrand monomer units at the surface.

In 1961 Michaels and Miekka[2] published a study of the reaction between poly(vinyl benzyl trimethylammonium) chloride and sodium polystyrenesulfonate (PSS) in which the results obtained were analogue to those of Fuoss and Sadek. Michaels also studied the process calorimetrically and found it to be virtually isoenthalpic.[3] He considered the obtained precipitates to be metastable reaction products[3-5] and explained the almost stoichiometric neutralisation with the geometrical compatibility of the reactants and the entropic favourability of the counterion release. In contrast with the experiments performed without simple electrolyte, the addition of the polyanion into a solution of polycation in the presence of NaBr (in both titrant and titrand) led to a large increase in particle size at lower ratios of monomers ($n_{-}/n_{+} = 0.6$). Moreover, the supernatant contained no polycation[2] and upon further addition of the polyanion the reaction proceeded very slowly. On the other hand, the suspensions prepared by titration of the polyanion with polycation were stabile until 1:1 monomolar ratio was reached and the addition of polycation above equivalent amount resulted in its slow incorporation in the precipitate. Michaels explained the described behaviour by differences in the conformation of the polycation (scrambled egg) and the polyanion (fully extended).[2]

In the early 70-thies Tsuchida[6] reported that soluble polyelectrolyte complexes can be formed by adding polyelectrolyte of low polymerization number (P_n) to a solution containing high P_n polyelectrolyte. Later on, Kabanov confirmed Tsuchida's results on several systems[7□-9] and was first to show that the obtained complexes were metastable reaction products; in so far as the solutions contained no simple electrolyte.[10-12]

For almost four decades Philipp and coworkers studied the interpolyelectrolyte neutralisation of high P_n polyelectrolytes with variable charge density.[13-16] The results of these investigations can be summarized as follows. The high charge density and high P_n polyions behaved identically to the system described by Fuoss and Sadek,[1] in that precipitates were formed from dispersions of approximately equimolar ratio of monomer units, while the addition of salt led to phase separation at lower r values. Philipp and co-workers explained the described behaviour by aggregation of colloid particles (Philipp used the term unstoichiometrical complexes[13,14]) induced by increase in the ionic strength. In contrast, the precipitates of low charge density polyelectrolytes deviated from equimolar monomer composition and the addition of electrolyte most often suppressed the complex formation.

The interest in reactions between oppositely charged polyelectrolytes increased rapidly after the introduction of polyelectrolyte multilayers (PEM) in the early nineties.[17] The reported results were usually consistent with earlier observations (formation of suspensions containing charged particles at lower ratios of monomer units,[18-20] and the formation of precipitates containing equivalent amounts of oppositely charged functional groups even at quite high concentrations of added electrolyte).[18] However, large deviations from 1:1 stoichiometry in reactions between high charge density and high P_n polyelectrolytes with no added electrolyte were reported[21] and even the fast formation of equilibrium products in systems containing high P_n polyions was occasionally suggested.[21,22] Unfortunately, the earlier efforts are nowadays often cited only as references of historical relevance, mainly due to the decline in the interest for synthetic polyelectrolytes during nineteen seventies and eighties. This has resulted with many repeated investigations and conclusions inconsistent with previous findings. Still there are many open questions to be answered. The most important one is certainly the time scale on which metastable products of interpolyelectrolyte neutralisation at surfaces and in solution exist under variable ionic conditions *(i.e* the time required for equilibrium establishment). Namely, this information is crucial for the successful application of polyelectrolyte assemblies, especially in biomedicine and biotechnology.[23-28] A particularly worthy step in that direction in the field of PEM research was made by Schlenoff and coworkers.[29-31] However, when working with such complex metastable materials as multilayers, the control of the conditions influencing their structure and composition can become demanding, not to mention the challenges of quantitative and qualitative analysis of nanocomposites. An answer to such a complicated problem presupposes the understanding of kinetics and thermodynamics governing the interpolyelectrolyte neutralisation in solution, especially at high electrolyte concentrations. In general, there are almost no reports regarding the chemical equilibrium position and the equilibrium product composition in reactions involving high charge density and high P_n polyelectrolytes in solution. As mentioned above, Fuoss[1] and Michaels[2–5] did not investigate the mentioned phenomena. The reports of Kabanov[10–12] on soluble, high charge density polyelectrolyte complexes suggest that the presence of electrolyte should be prerogative for formation of equilibrium products in systems containing high P_n polyions. In accord, Dautzenberg[18] reported that the equilibrium in solution containing polydiallyamonium chloride and sodium polystyrenesulfonate can be reached only in presence of NaCl. The equilibrium product was virtually insoluble polysalt even at quite high concentrations of salt. On the other hand, the structural rearrangements in some metastable precipitates could be much faster. Interestingly, Michaels[2] noticed the asymmetric composition of equilibrium products of high charge density polyelectrolytes at molar ratios different that 1:1, irrespectively of the addition order. However, he did not perform detailed investigations, so that the phenomenon remained unexplored.

The contemporary investigations of PEMs have also indicated that the influence of electrolyte concentration on the size and the composition of reaction products should be more thoroughly investigated. By taking into account the results of Tsuchida[6] and Kabanov,[7-12] and those of Michaels[2-5] and Dautzenberg,[18,19] the increase in electrolyte concentration could lead either to aggregation of colloid particles (the particularly favoured explanation), faster establishment of equilibrium and/or asymmetric composition of the metastable and even equilibrium reaction products (mentioned explicitly only by Michaels[2]). Since the increase in the thickness of multilayers[32-35] accompanied with incorporation of counterions within the nanocomposites[36,37] was noticed at higher electrolyte concentrations, the aggregation of colloid particles without the changes in the conformation of polyions and, most likely, the composition of products (*i.e.* the compensation of unpaired monomers with simple counterions) can in many cases be excluded. The recent investigations of interpolyelectrolyte neutralisation in solution by Chen et al.[38,39] support these statements. However, the phase separation, induced by electrolyte addition, is still most frequently attributed only to aggregation of primary complexes.

It should also be noted that reversible swelling[40] and ion-specific counterion binding within the multilayers[41] were observed upon exposing some of the nanocomposites to concentrated salt solutions, in particular those prepared from polyelectrolyte solutions containing relatively high salt concentrations. Furthermore, the strong influence of the electrolyte type on the composition and thickness of multilayers at higher ionic strengths was observed.[42-45] The authors ascribed these findings to differences in the energetics of counterion binding to polymer chains. However, it is interesting to note that the multilayer build-up becomes ion-specific only at quite high electrolyte concentrations. This fact is difficult to explain solely by the differences in the enthalpies of counterion removal, since it indicates that the ion specificity should be present even at low salt concentrations. Similar ion-specific effects could be expected in reactions taking place in solutions, where the phenomenon remained virtually unexplored. Moreover, the strongly ion-specific built-up of polyelectrolyte multilayers can be largely understood on the basis of corresponding processes in solution. This opens a path for simple modifications of their structure and composition by variations in electrolyte concentration and type.

Generally, the formation of polyelectrolyte assemblies (complexes and multilayers) is usually explained by the entropy gain resulting from the release of counterions. [1,3,18] Although some of the reactions were found to be isoenthalpic,[3,21,22,46] the interpolyelectrolyte neutralisation has hardly been investigated calorimetrically. The calorimetric data regarding the interpolyelectrolyte neutralisation in solution could be of considerable importance to researchers concerned with energetics of multilayer formation, overcharging in particular.[47,48] Namely, the formation of charged polyelectrolyte complexes (particles) and neutral precipitates in solution can be studied by titration calorimetry. Moreover, the influence of the electrolyte type and concentration on the reaction thermodynamics can be rationalized on the basis of calorimetric measurements, at least on a semi-quantitative level.

With regard to some of the unresolved questions concerning the interpolyelectrolyte neutralisation in solution, we have decided to systematically study the complexation between the polyallylammonium cation and polystyrenesulfonate anion in the presence of electrolytes containing the sodium cation and different monovalent anions (X^-; X = F, Cl, Br, I, NO_3, ClO_4) by means of microcalorimetry, DLS, electrokinetics and spectrophotometry. The main goal of the research was to investigate the reaction thermodynamics and the composition of equilibrium and metastable reaction products, particularly stressing the role of electrolyte concentration and counteranion type. The polycation and polyanion were chosen because of their frequent use in the process of polyelectrolyte multilayer formation. Recently, those nano-composites have been proposed as biocompatible coatings which prevent the formation of cloths in artificial blood vessels and stents.[49,50] In another investigation they have been used as surfaces for vascular cell growth around synthetic implants.[49,51] The interest in determining their long term stability and the possibility of preparing PEMs containing higher amounts of polycations than of polyanions (the cells adhere to positively charged PAH) is hence considerable. The choice of the anions was made in accordance with the differences in their size, structure and the related hydration. The obtained results have provided deeper insight in the process of interpolyelectrolyte neutralisation of the investigated polyions in solution and at surfaces which enables aimed modifications of PAH/PSS nanocomposites by variation of ionic conditions.

2. Experimental

2.1 *Materials*

Polyallylamine hydrochloride (PAHCl) (M_w = 70000 g mol^{-1}) was purchased from Aldrich. The degree of monomer functionalisation (f) was determined by potentiometric titration of PAH samples with standardized solution of sodium hydroxide. The value obtained was $f = 0.94$.

Polysodium 4–styrenesulfonate, NaPSS (M_w = 77000 g mol^{-1}) analytical standard for GPC was obtained from Aldrich and was used as received.

The sodium salts of monovalent anions (NaCl, NaBr, NaI, $NaNO_3$, $NaClO_4$,) were obtained from various manufacturers (Merck, Sigma, Aldrich) and were of analytical purity grade. The solutions were prepared by dissolving the polyelectrolyte and simple electrolyte in aqueous solution of hydrochloride acid (c(HCl) / mol dm^{-3} = 1×10^{-3} or 1×10^{-4}). The acid was added to keep the polycation monomers protonated.[52]

2.2 *Dynamic light scattering and electrokinetic measurements*

The complexes of polystyrenesulfonate anion with pollyallyammonium cation were prepared by titration of the solution of one polyelectrolyte with the solution of the other. In order to avoid the dependence of particle size upon stirring rate[13] the titrant was added under vigorous stirring. The effective hydrodynamic radii (D_eff) of the accordingly prepared complexes were determined from dynamic light scattering experiments. For this purpose a *Brookhaven 90 Plus Particle Size Analyzer* (*Brookhaven Instruments Corporation*) was used. The detector was in all cases placed at 90° with respect to the incident beam. The diffusion coefficients of particles (d) were calculated from the autocorrelation function and D_h of particles were obtained from the Einstein-Stokes equation:[53,54]

$$D_\mathrm{eff} = \frac{k_\mathrm{B}T}{3\pi\eta d}, \qquad (2.2.1)$$

where k_B denotes the Boltzmann constant, T thermodynamic temperature and η the medium (water) viscosity.

The electrophoretic mobilities of the complexes were determined by means of *ZetaPlus* (*Brookhaven Instruments Corporation*) from the Doppler shift frequency ($\Delta\omega$) and the applied electric field (E):[53,54]

$$\Delta\omega = \left(\frac{2\pi n}{\lambda}\right) Eu \sin\theta. \qquad (2.2.2)$$

In the above equation, n and u are the medium refractive index and electrophoretic mobility of the particles, respectively. The scattering angle during measurement was fixed (θ = 15°). The temperature was regulated with an internal thermostatting device, and measurements were made at 25 °C.

2.3 *Calorimetric investigations*

Microcalorimetric titrations were carried out by means of isothermal titration microcalorimeter, CSC 4200 ITC (*Calorimetry Science Corporation*) at 25.0 ± 0.1 °C. The calorimeter reaction cell was filled with polycation or polyanion solution ($V = 1.3\ \mathrm{cm}^3$; c_m(titrand) / mol dm^{-3} ≈ 5×10^{-3} or 2×10^{-2}) which in some experiments contained simple electrolytes. The enthalpy changes were recorded upon stepwise additions (6 min intervals) of oppositely charged polyelectrolyte (c_m / mol dm^{-3} ≈ 3×10^{-1} or 7.5×10^{-2}) under the same ionic conditions as in the reaction cell from a 250 μL Hamilton syringe. As in the case of DLS experiments, the stirring rate was kept high (300 rpm) in order to avoid the dependence of particle size upon stirring rate. Blank experiments were performed in order to make corrections for the enthalpy changes corresponding to dilution of titrant. The heat effect of titrand dilution was negligible in all cases.

2.4 *Spectrophotometric measurements*

The amount of polystyrenesulfonate in the supernatant obtained above the molar ratios at which flocculation occurred (usually at 1:1 molar ratio of oppositely charged monomer units) was determined spectrophotometrically by means of *Varian Cary 5* spectrophotometer at 25.0 ± 0.1. In a typical experiment the total amount and concentration of titrant and titrand were held constant. The suspensions were prepared by titration of one reactant with the other under vigorous stirring. When the desired molar ratio was reached, the samples were centrifuged and the aliquots of supernatant solutions were taken for UV measurements. To test the reversibility of complex formation titrations up to 1:1 molar ratio of monomer units were run in both directions.

3. Results and discussion

3.1 *Complexation between polyallylammonium anion and polystyrenesulfonate cation without added supporting electrolyte.*

The reaction between polyallylammonium cation and polystyrenesulfonate anion was studied by means of microcalorimetry, DLS, electrokinetics and spectrophotometry in order to characterize the reaction products and to investigate the corresponding reaction energetics. In the case of interpolyelectrolyte neutralisation of sodium polystyrenesulfonate with polyallylammonium chloride without added electrolyte the charge of products was investigated electrokinetically.

The results of electrophoretic measurements, obtained by titration of polycation with polyanion (Figure 1. a)) and polycation with polyanion (Figure 1. b)), indicate that at lower molar ratios of monomers, charged colloid particles are being formed (the *n*(PSS) / *n*(PAH) at abscissa stands for molar ratio of negative and positive monomers). Evidently, the sign of their charge is equal to that of the titrand polyions.

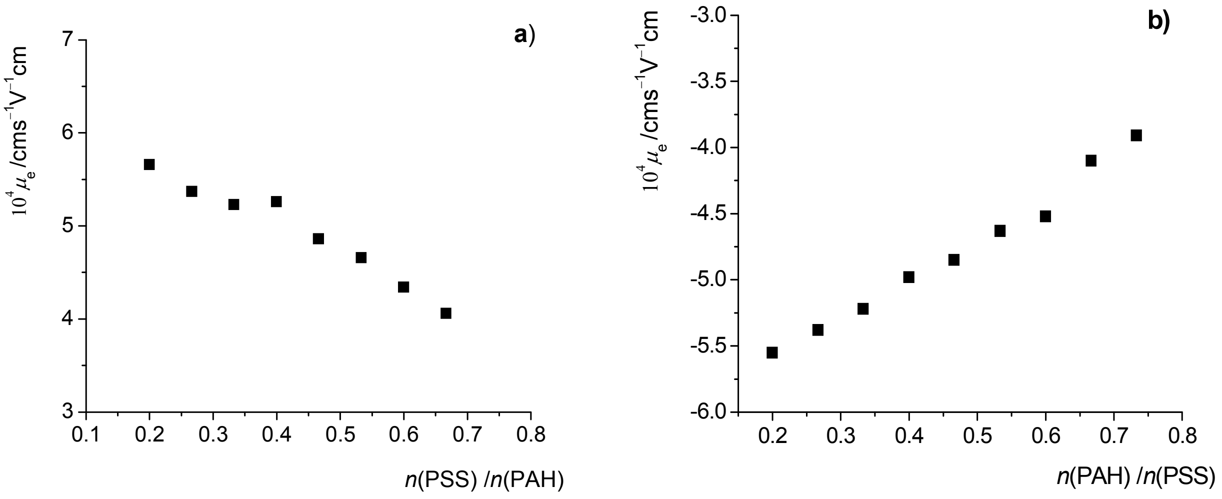

Figure 1.a) Electrophoretic mobility of colloid complexes obtained by titration of polyallylammonium chloride (c_m = 5×10^{-3} mol dm^{-3}, V_0 = 1.5 mL) with sodium polystyrenesulfonate (c_m = mol dm^{-3}) at 25 ºC.

b) Electrophoretic mobility of colloid complexes obtained by titration of sodium polystyrenesulfonate (c_m = 5×10^{-3} mol dm^{-3}, V_0 = 1.5 mL) with polyallylammonium chloride (c_m = mol dm^{-3}) at 25 ºC.

The size of the particles (Figure 2) is dependent on the reactant concentration, *i.e.* the particles do not have a fixed composition. They are merely metastable products of the reaction in which an insoluble product comprised of oppositely charged polyelectrolytes is formed. As is customary in the literature, these particles will be referred to as (colloid) polyelectrolyte complexes. It can also be seen that the size of the complexes remains almost constant in a wide range of monomer unit ratios and then suddenly increases (flocculation). Obviously, the predominant process taking place at lower molar ratios is the formation of new particles and not the neutralisation of charged groups at the surface of already present products.

The onset of flocculation is visible both from electrophoretic and DLS experiments (the particle size suddenly becomes too large for both techniques). The flocculation in all cases (except at lowest concentration of reactants) occurs before the equivalent amount of titrant is added (close to monomer ratio (r = n(PSS) / n(PAH) or r = n(PAH) / n(PSS) equal to 0.8). However, a clear shift of flocculation to lower r values was noticed upon the increase in concentration of reactants.

The interpolyelectrolyte neutralisation of the investigated polyelectrolytes hence proceeds as suggested by Fuoss and Sadek.[1] The process initially results in formation of charged primary complexes. (As noted before, these products have often been referred to as unstoichiometrical[13] or semisoluble[55] complexes.) Due to fast, random interpolyelectrolyte neutralisation and the large polymerisation numbers of both reactants the primary complexes bear a surplus of titrand groups at the surface. Consequently, the neutralisation of "dangling chains",[1] accompanied by a sudden increase in particle size (formation of secondary particles whose charge is of the opposite sign that that of primary), has to take place before added equivalent amount of titrant monomer units, since completely free titrand chains no longer exist in solution. In the end, due to geometric compatibility of polyions[4] the reaction comes to a halt at an approximately 1:1 monomer molar ratio, when most of the oppositely charged groups become paired.

It should be mentioned that the size of particles was independent on the rate of reactant addition, but was dependant on the stirring rate.[15] However, the mentioned dependence could be noticed only at extremely low stirring rates. In order to avoid this phenomenon the reaction mixtures were stirred vigorously during titrans addition. The obtained D_{eff} were also quite reproducible (within ± 20 %). This can be clearly seen from the similar values of particle size obtained in the low salt regime (0.1 mol dm^{-3} NaX) irrespectively of the electrolyte type.

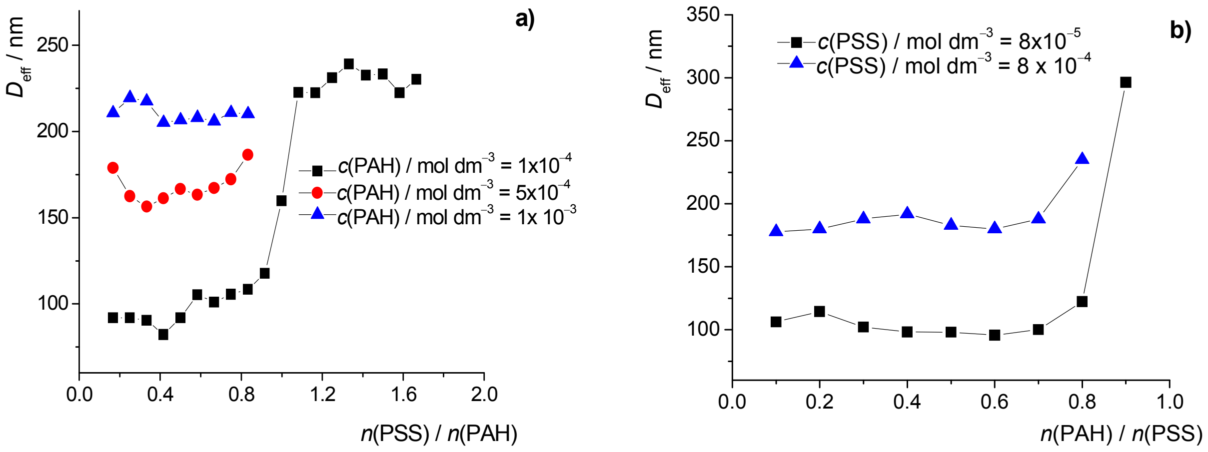

Figure 2. a) Hydrodynamic diameters of colloid complexes obtained by titration of polyallylammonium chloride (V_0 = 2.0 mL with sodium polystyrenesulfonate at 25 ºC. Concentration of titrand is given in Figure. The titrant concentration all cases equalled 20× $c_m(\mathrm{PAH}_{70})$.

b) Hydrodynamic diameters of colloid complexes obtained by titration of sodium polystyrenesulfonate (V_0 = 2 mL) with polyallylammonium chloride at 25 ºC. Concentration of titrand is given in Figure. The titrant concentration all cases equalled 20× $c_m(\mathrm{PAH}_{70})$.

This is clearly seen from the hydrodynamic diameters of particles obtained by titration of the polyanion with the polycation at the lowest concentration of reactants (Figure 2. a)). In the mentioned experiment flocculation results in secondary complexes whose size is almost constant above the 1:1 molar ratio. The high degree of monomer pairing was independently confirmed by calorimetry and by spectrophotometry (the supernatant at 1:1 molar ratio contained only traces of NaPSS irrespective of the polyelectrolyte addition order). It should be noted that all monomer units of opposite charge cannot be paired, due to different functionalisation degrees (*f*) of the polyions. However, the functionalisation degree of the polycation is large enough that its deviation from unity probably does not affect the pairing of charged groups (product structure).

Despite the apparent reversibility of the process (*i.e.* the flocculates have a very similar chemical composition, but have different structure), the fast interpolyelectrolyte neutralisation results in metastable products. This is most evident from the results of spectrophotometric measurements at higher electrolyte concentrations (chapter 3.3). It should also be mentioned that, the investigations carried out so far of reactions involving strong polyelectrolytes revealed the equilibrium could be reached only in the presence of a simple electrolyte.[10-12,18] Therefore, the fast formation of the equilibrium product without added electrolyte in the case of the macromolecules investigated herein seems highly unlikely.

In contrary to earlier reports[21] the described results suggest that no homogeneous equilibrium (*i.e.* soluble complexes in equilibrium with the reactants) exists in the described system. Due to high P_n numbers of both reactants, only the heterogeneous equilibrium is possible[6-12] (*i.e.* insoluble precipitate in equilibrium with free polyelectrolytes in solution). The structure of the formed precipitate should be comprised of sequences which are neutralised not with one but with several oppositely charged polyions[3,4]. The TEM photography is in accord with such a hypothesis (supporting information, Figure S1).

The thermodynamics governing the interpolyelectrolyte neutralisation of PAH and PSS can be deduced on the basis of calorimetric experiments. The cumulative enthalpy changes obtained by titrations of polycation with polyanion and *vice versa* are shown in Figure 3. a) and b)). It should be noted that no dependence of measured enthalpy changes upon stirring rate or the rate of titrans addition could be observed (supporting information, Figure S2). Such results indicate that the quite low stirring rates (50 rpm), in comparison to those usual in titration calorimetry (300 rpm or higher), are sufficient for effective mixing of reactant solutions (preparation of primary polyelectrolyte complexes).

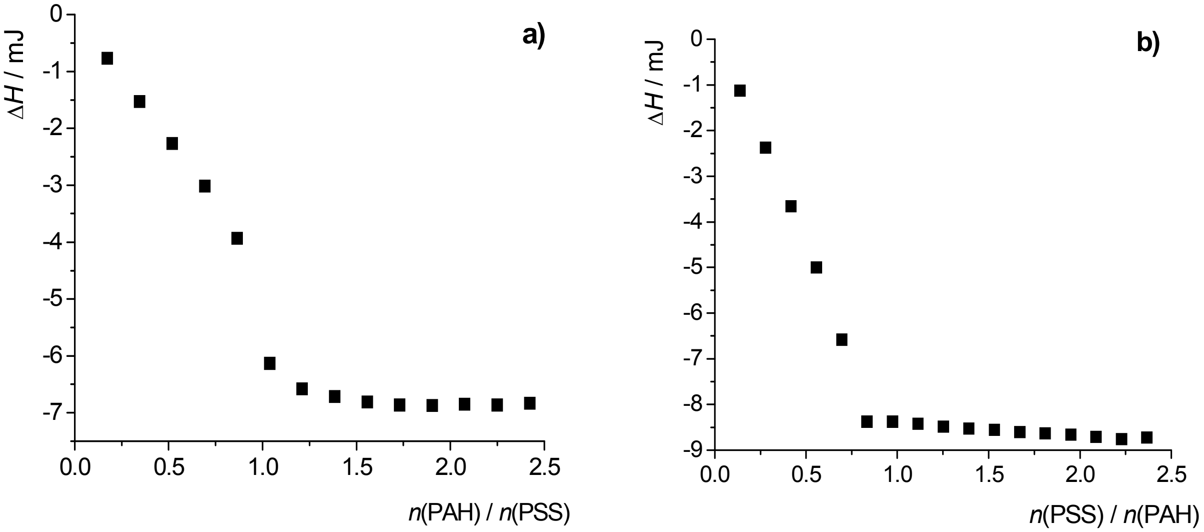

Figure 3. **a)** Cumulative enthalpy changes obtained by titration of polyallylammonium chloride ($c_\mathrm{m} = 5\times10^{-3}$ mol dm^{-3}, $V_0 = 1.3$ mL) with sodium polystyrenesulfonate ($c_\mathrm{m} = 6\times10^{-2}$ mol dm^{-3}), $\theta = (25.0 \pm 0.1)$ ºC.
b) Cumulative enthalpy changes obtained by titration of sodium polystyrenesulfonate ($c_\mathrm{m} = 5\times10^{-3}$ mol dm^{-3}, $V_0 = 1.3$ mL) with polyallylammonium chloride ($c_\mathrm{m} = 7.5\times10^{-2}$ mol dm^{-3}), $\theta = (25.0 \pm 0.1)$ ºC.

As it can be seen, the interpolyelectrolyte neutralisation resulted in measurable negative enthalpy changes, irrespectively of the addition order. The differences in the measured trend of ΔH between the two types of experiment are clearly visible. In the case of titration of PAH with PSS no enthalpy changes above n(PSS) / n(PAH) = 0.75 can be observed. In contrast, measurable enthalpy changes for titration of PSS with PAH were noticed up to n(PAH) / n(PSS) = 1 (or even slightly higher). These results confirm that, depending on the order of reactant addition, different primary complexes are being formed.

By taking into account the results of DLS and calorimetric measurements one can conclude that changes in the trend of cumulative ΔH values in Figures 3 a) and b) occur at the onset of flocculation. The neutralisation of the corona is therefore less enthalpically favourable than the formation of positive colloid complexes, while the opposite holds for negative complexes. The somewhat surprising fact is that both the neutralisation of "dangling chains" and the formation of primary particles are in essence isoenthalpic. This becomes clear once the measured enthalpy values before the flocculation are divided with the total amount of added titrant monomer units. Namely, due to almost complete neutralisation of titrant charged groups in the core (the functionalisation degrees of both polymers are close to unity), these values can be considered as the enthalpies of formation of colloid complexes according to the following reaction:

$$x\mathrm{PAH} + \mathrm{PSS} \rightarrow \mathrm{PSS(PAH)}x, \qquad (3.1.1)$$

where $x > 1$ (the surplus of PAH monomer units ate the surface of complexes). By analogy, the reaction for the formation of negative complexes will be given by the expression:

$$y\mathrm{PSS} + \mathrm{PAH} \rightarrow -\mathrm{PAH(PSS)}y, \qquad (3.1.2)$$

with $y > 1$. The values calculated for both type of complexes are enlisted in Table 1.

Table 1. Complexation thermodynamics of sodium polystyrenesulfonate with polyallylammonium chloride; enthalpies of formation of charged colloid complexes; $\theta = (25.0 \pm 0.1)$ ºC.

$(\Delta_c H_{(\mathrm{PSS} \to \mathrm{PAH})} \pm \mathrm{SE})$ / kJ mol^{-1}	$(\Delta_c H_{(\mathrm{PAH} \to \mathrm{PSS})} \pm \mathrm{SE})$ / kJ mol^{-1}
-1.48 ± 0.05	-0.80 ± 0.05

Evidently, the complexation enthalpies are quite close to zero. The difference between $\Delta_c H_{(\mathrm{PSS} \to \mathrm{PAH})}$ and $\Delta_c H_{(\mathrm{PAH} \to \mathrm{PSS})}$ is thermodynamically irrelevant, though the enthalpy of formation of positive complexes is almost twice as large (absolute value) as that of negative complexes. In fact, $\Delta_c H$ for both directions are so close to zero that they could only be determined precisely because a quite sensitive microcalorimeter was used.

The almost isoenthalpic formation of colloid complexes and the neutralisation of the negative and positive groups at the surface of primary complexes is strong evidence that the overcharging in the process of multilayer formation is not as energetically demanding as sometimes stressed in the literature.[47,48] It is sufficient to imagine the long, dangling chains at the surface of a neutralised core of a colloid complex to explain the notable "overcharging" of the multilayer surface. The protruding chains, if sufficiently far apart, could be similar (in terms of counterion condensation) to free chains in solution. This explanation, proposed by Fuoss[1] for polyelectrolyte complexes and applied by Schlenoff for multilayers,[56] can account both for notable increase in the multilayer thickness at each deposition cycle as well as isoenthalpic formation of primary complexes and the neutralisation of the corona. On the other hand, it could be that fast complexation leads to repulsion of the kinetically trapped corona chains which triggers further counterion condensation. Such possibility needs to be investigated further, for instance by monitoring the activity of counterions released during formation of primary and secondary complexes.

Due to the large amount of paired monomer units in the final products (precipitates) their formation must be isoenthalpic as well. The reaction enthalpies for precipitate formation, (expressed per monomer of titrand) amount to −1.3 ± 0.1 kJ mol^{-1} (PSS→PAH) and −0.89 ± 0.09 kJ mol^{-1} (PAH→PSS). The given values should be close to the enthalpy of polyelectrolyte salt (polysalt) formation since no long-range order can be expected in the polysalt structure.[4]

One should be aware that although almost complete neutralisation of monomer units in the core of colloid complexes takes place, the amount of neutralised titrand functional groups at the surface of primary complexes (corona) could be lower. Likewise, the amount of titrant needed for neutralisation of the corona charge might also be somewhat higher than 1:1. This can lead to flocculate "overcharging".

Since the reaction of PSS with PAH is virtually isoenthalpic, the process is almost entirely governed by the positive entropy changes as a result of counterion release. The entropically favourable expulsion of counterions was suggested to be a major driving force in interpolyelectrolyte neutralisation already by Fuoss and Sadek.[1] The first experimental confirmation for this hypothesis was provided by Michaels.[3] The recent calorimetric study of the reaction of sodium polystyrensulfonate and with polydiallyldimethylammonium chloride (PDADMACCl) revealed that the mentioned process was isoenthalpic as well.[22] One of the most important findings for the understanding of thermodynamics governing the interpolyelectrolyte neutralisation of highly charged polyions, is that the reaction of NaPSS and PDADMACCl remained quantitative at 25 °C (all oppositely charged groups present at equimolar ratio form polysalt even after the equilibrium had been reached, even at quite high NaCl concentrations[18]). By analogy, due to high charge density of PAH and PSS their interpolyelectrolyte neutralisation must be strongly entropically favoured, and hence quantitative as well. (See section 3.3.) This should hold for all isoenthalpic reactions involving high charge density polyelectrolytes. However, by no means must the enthalpies of all such reactions be close to zero, as was occasionally suggested.

3.2 *Complexation between polyallylammonium anion and polystyrenesulfonate cation in NaCl(aq)*

The complexation of PSS and PAH was also studied in the presence of NaCl. The solutions of both reactants in all cases contained equal concentrations of simple electrolyte. The results of DLS investigations, performed by running the experiments in both directions, are presented in

Figure 4. a) and b). As it can be seen, the increase in NaCl concentration results in a large shift in the flocculation onset toward lower monomer molar ratios in the case when polyanion was added to the solution of polycation. Quite opposite, the sudden increase in the particle size in the case of titration in the opposite direction occurs at similar molar ratio, irrespective of the electrolyte concentration. This is yet another proof that oppositely charged colloid complexes are being formed. The described results also suggest that the studied polyelectrolytes have different distribution of counterions in the double layer (the increase in particle size would otherwise occur at similar molar ratios in both directions). Clearly, the chlorides are more strongly condensed to PAH than the sodium ions are to PSS chains. The negatively-charged corona keeps the colloids stable, while positive does not.

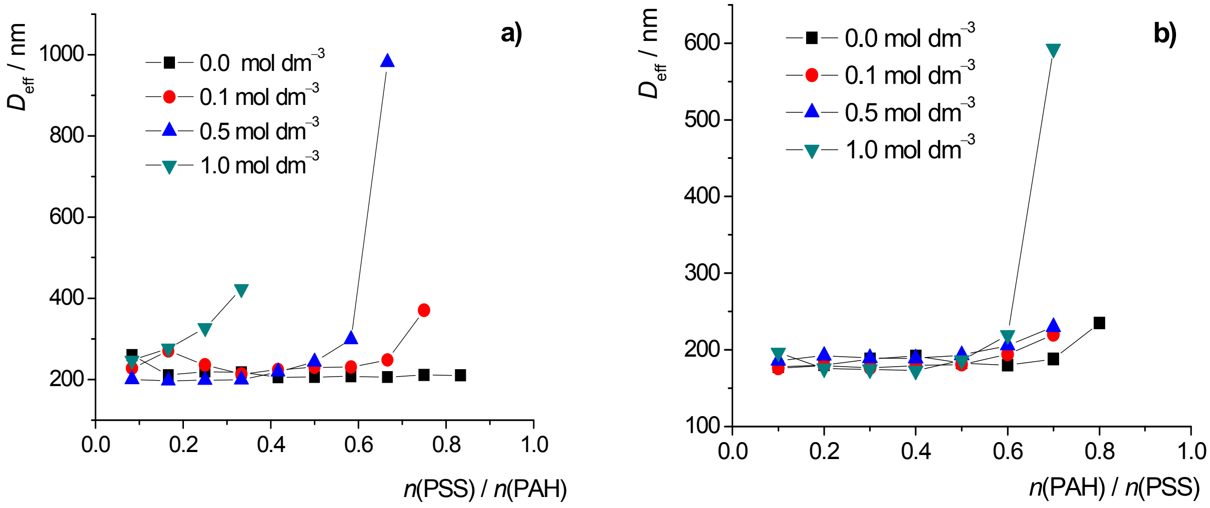

Figure 4. **a)** Hydrodynamic diameters of colloid complexes obtained by titration of polyallylammonium cation (c_m $=1\times10^{-3}$ mol dm^{-3}, $V = 2$ mL) with polystyrenesulfonate anion ($c_m = 1.7\times10^{-2}$ mol dm^{-3}) in NaCl(aq) at 25 ºC.
b) Hydrodynamic diameters of colloid complexes obtained by titration of polystyrenesulfonate ($c_m = 1\times10^{-3}$ mol dm^{-3}, V = 2 mL) with polyallylammonium cation ($c_m = 2\times10^{-2}$ mol dm^{-3}) in NaCl(aq) at 25 ºC.

The sudden increase in particle size at low ratios of monomer units for PSS→PAH experiments at higher ionic strength can be explained by aggregation of the primary positive colloid complexes. To test the proposed rationale, a solution of PAHCl was titrated with NaPSS up to n_{-}/n_{+} = 0.5. (the solutions of both reactants did not contain NaCl). To the accordingly prepared suspensions NaCl(aq) was slowly added and the particle size was monitored. The results are shown in Figure 5. Obviously, the aggregation of primary complexes takes place at concentrations of NaCl higher than 0.5 mol dm^{-3}. However, although the aggregation of positive complexes during the experiments in which NaCl was added to solutions of both reactants (Figure 4.a) most certainly occurs, other effects must be considered as well. One should keep in mind the difference in counterion distribution can result with less extended conformation of negative, and to a much larger extent of positive chains in the case when NaCl is added to titrant and titrand solutions. This can lead to partial interpolyelectrolyte neutralisation in the core (Michaels[2]) and, most likely, the more pronounced overcharging of positive with respect to negative complexes.

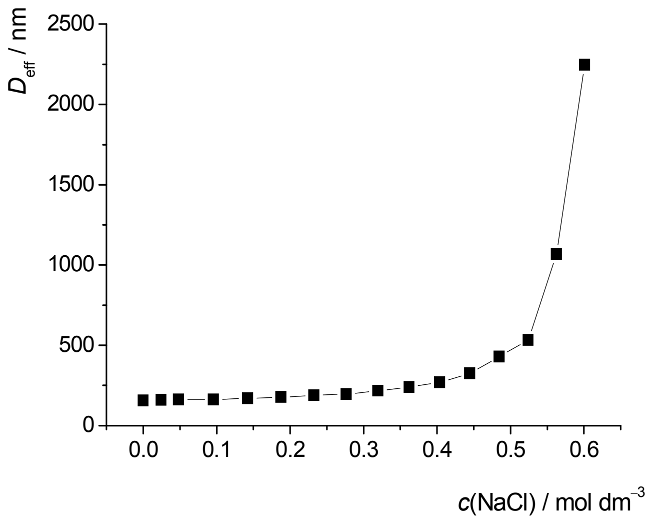

Figure 5. Hydrodynamic diameters of particles obtained by titration of positive colloid complexes (prepared by addition of sodium polystyrenesulfonate (c_m = 2 ×10^{-2} mol dm^{-}3) to solution of polyallylammonium chloride (c_m = 1 ×10^{-3} mol dm^{-3}, V = 2 mL); n(PSS) / n(PAH) = 0.5)) with NaCl (c = 5 mol dm^{-3}) at 25 ºC.

The primary complexes prepared with and without added NaCl, hence do not need to contain similar ratio of paired groups, or surplus of monomer units in the corona. On the other hand, both of them would be prone to aggregation, as evident from the previously described experiment (Figure 5). A more detailed discussion on the subject will be given in the following section. For now, it should be noted that the results of spectrophotometric measurements suggest that the majority of monomer units in the precipitates are paired in the studied range of NaCl concentrations (Figure S3, supporting information). The spectra of supernatants (obtained by titrations run in both directions) even at 1 mol dm^{-3} showed no traces of free NaPSS.

The interpolyelectrolyte neutralisation of PAH with PSS was also studied calorimetrically in the same range of NaCl concentrations. The thermograms for titrations of polycation with polyanion and the cumulative enthalpy changes, recorded during titrations in both direction (c(NaCl) = 0.1 mol dm^{-3}), are given in supporting information (Figures S4 and S5). The calculated enthalpies for formation of positively and negative colloid complexes can be found in Table 2. Somewhat surprisingly, the enlisted values suggest that the increase in electrolyte concentration has very little effect on the energetics of interpolyelectrolyte neutralisation. This finding can be reconciled with observed aggregation of the positive complexes only if different distributions of counterions around the polyion at high ionic strength and with no added salt result in similar changes in enthalpy associated with removal of the corresponding ionic atmospheres (including both the directly and indirectly bound counterions).

Table 2. Complexation thermodynamics of polystyrenesulfonate anion with polyallylammonium cation; enthalpies of formation of charged colloid complexes in NaCl(aq); θ = (25.0 ± 0.1) ºC.

c(NaCl) / mol dm^{-3}	($\Delta_c H_{(PSS \rightarrow PAH)}$ ± SE) / kJ mol^{-1}	($\Delta_c H_{(PAH \rightarrow PSS)}$ ± SE) / kJ mol^{-1}
0.01	−1.36 ± 0.04	[a]
0.05	−1.74 ± 0.01	[a]
0.1	−2.02 ± 0.02	−1.74 ± 0.09
0.5	−2.6 ± 0.1	−1.76 ± 0.06

[a] the interpolyelectrolyte neutralisation of PSS with PAH was not investigated at this NaCl concentration.

As mentioned in the chapter 4.3.1 the results of Kabanov[7–9] and Dautzenberg[18] indicate that the presence of a simple electrolyte in solution is a prerogative for equilibrium. However, the fast establishment of equilibrium in the case of NaCl addition in suspensions containing negative colloid complexes can be ruled out. The negative particles prepared in 1 mol dm^{-3} NaCl remained stable for months. The fast formation of equilibrium products once the flocculation or aggregation of positive complexes occurred can also be rejected on the basis of results obtained in concentrated $NaClO_4$ and $NaNO_3$ solutions, which will be described in the following section.

3.3 *Complexation between polyallylammonium anion and polystyrenesulfonate cation in NaX(aq)*

The investigations of interpolyelectrolyte neutralisation in NaX(aq) solutions were performed at identical concentrations of monomer units as described previously. The concentration of NaX was always kept equal or higher than 0.1 mol dm^{-3} in order to assure the complete substitution of chlorides on PAH chains with the anions of added electrolyte ($c(\text{X}^{-})$ was always at least 20 times higher than that of chlorides which counterbalanced the PAH charge). The NaX concentration was in all cases equal in both titrant and titrand solutions.

The DLS results obtained in 0.1 mol dm^{-3} NaX (Figure 6.) reveal quite weak ion-specific effects. The reaction in NaBr(aq) and NaI(aq) proceeds as in aqueous solution of NaCl. By contrast, in the case of $NaNO_3$ and $NaClO_4$ the sudden increase in particle size always occurred at lower ratios of monomer units (roughly 10 %). Despite the mentioned differences, the particle size measurements indicate that the amount of paired groups in all precipitates should still be high. This was confirmed by spectrophotometric measurements. The supernatant spectra at 1:1 molar ratio of monomer units contained no NaPSS, irrespective of the addition order in all investigated electrolyte solutions (supporting information, Figures S6 and S7). Note that spectrophotometric measurements could not be performed in $NaNO_3$(aq) and NaI(aq) due to the strong absorption of nitrate and triodide ion.

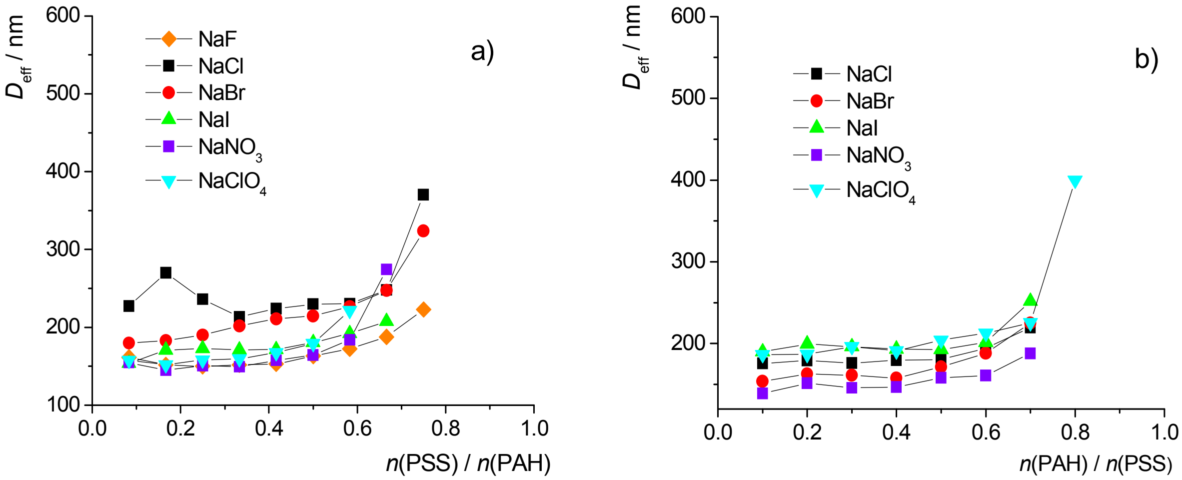

Figure 6.a) Hydrodynamic diameters of colloid complexes obtained by titration of polyallylammonium cation (c_m $=1\times10^{-3}$ mol dm^{-3}, $V_0 = 2$ mL) with polystyrenesulfonate anion ($c_\mathrm{m} = 1.7\times10^{-2}$ mol dm^{-3}) in NaX (aq, $c = 0.1$ mol dm^{-3}) at 25 ºC.

b) Hydrodynamic diameters of colloid complexes obtained by titration of polystyrenesulfonate anion (c_m $=1\times10^{-3}$ mol dm^{-3}, $V = 2$ mL) with polyallylammonium cation ($c_\mathrm{m} = 2.0\times10^{-2}$ mol dm^{-3}) in NaX (aq, $c = 0.1$ mol dm^{-3}) at 25 ºC.

The interpolyelectrolyte neutralisation was also studied calorimetrically. Thermograms recorded by titration of polycation with polyanion in NAX (aq, c = 0.1 mol dm^{-3}) are shown in Figure 7. The corresponding cumulative and successive enthalpy changes obtained in NaF and $NaClO_4$ are given in Figures 8. a) and b).

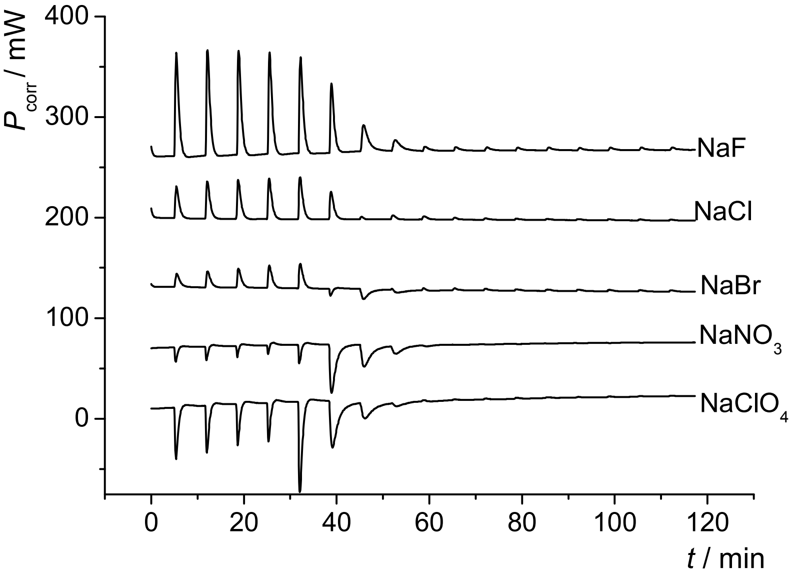

Figure 7. Thermograms obtained by titration of polyallylammonium cation ($c_\mathrm{m} = 5\times10^{-3}$ mol dm^{-3}, $V = 1.3$ mL) with polystyrenesulfonate anion ($c_\mathrm{m} = 6\times10^{-2}$ mol dm^{-3}) in NaX(aq, ($c = 0.1$ mol dm^{-3}), $\theta = (25.0 \pm 0.1)$ º C.

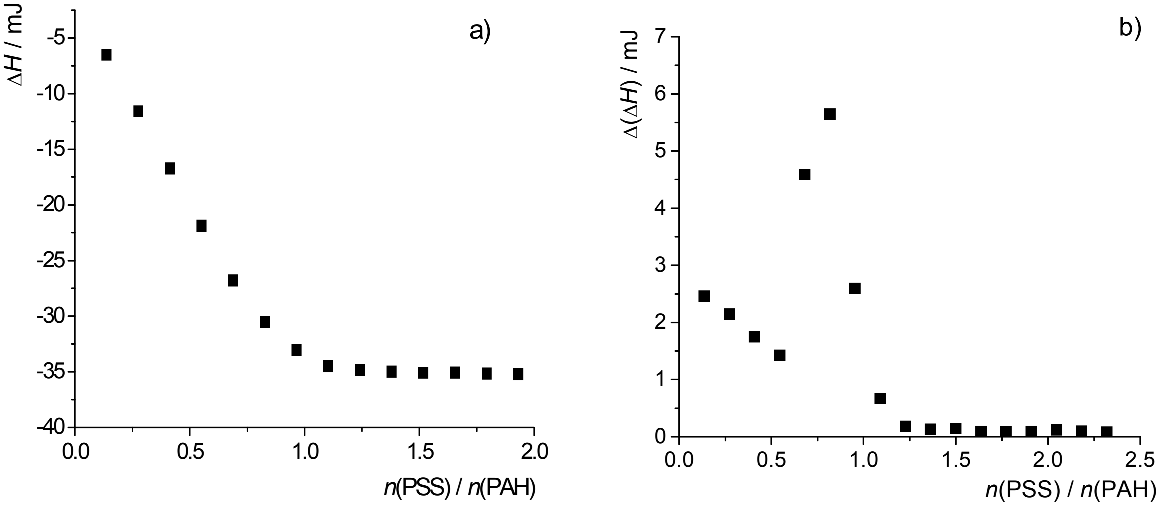

Figure 8.a) Cumulative enthalpy changes obtained by titration of polyallylammonium cation (c_m = 5.0×10^{-3} mol dm^{-3}, V_0 = 1.3 mL) with polystyrenesulfonate anion (c_m = 6.0×10^{-2} mol dm^{-3}) in NaF (c = 0.1 mol dm^{-3}), θ = (25.0 ± 0.1) ºC. **b)** Successive enthalpy changes obtained by titration of polyallylammonium cation (c_m = 5.0×10^{-3} mol dm^{-3}, V_0 = 1.3 mL) with polystyrenesulfonate anion (c_m = 6.0×10^{-2} mol dm^{-3}) in NaClO$_4$ (c = 0.1 mol dm^{-3}), θ = (25.0 ± 0.1) ºC.

By examining the presented data one can see that the onset of flocculation is accompanied by quite distinct changes in the trend of ΔH. The values are in all cases less favourable than at lower molar ratios of monomer units, at which colloid complexes are formed. This could be due to the higher amount of condensed counterions at the corona with respect to free chains in solution. In contrast, the interpolyelectrolyte neutralisation which occurs during flocculation can lead to a lower amount of paired monomers (previous section), although thermodynamically, the differences are not large. This is evident from the relatively low enthalpies of formation of the positive colloid complexes (Table 3, column 2).

Obviously, the complexation process is in most cases entirely entropically driven (positive complexation enthalpies). However, the difference between the most favourable and most unfavourable complexation enthalpies amounts to almost 8 kJ mol^{-1}, which is not negligible. From the enthalpic point of view, the polysalt should be most stable in concentrated solutions of NaF. Its formation is enthalpically most demanding in NaClO$_4$. Interestingly, the complexation enthalpies can be correlated with standard hydration enthalpies of the counteranions.[57] (Table 3, column 4)- The more negative the enthalpy of anion hydration is, the more energetically demanding the complexation becomes. A similar correlation was previously observed for monovalent cation binding to the polystyrenesulfonate group.[58]

Table 3. Complexation thermodynamics of polystyrenesulfonate anion with polyallylammonium cation; enthalpies of formation of charged colloid complexes in NaX (c = 0,1 mol dm^{-3}); $\theta = (25.0 \pm 0.1)$ °C.

NaX	$(\Delta_c H_{\mathrm{PSS} \to \mathrm{PAH}} \pm \mathrm{SE})$/ kJ mol^{-1}	$(\Delta_c H_{\mathrm{PAH} \to \mathrm{PSS}} \pm \mathrm{SE})$/ kJ mol^{-1}	$\Delta_{\mathrm{hyd}} H^{\circ}(\mathrm{X}^{-})$ / kJ mol^{-1}
NaF	-5.60 ± 0.05	-5.72 ± 0.06	-510
NaBr	≈ 0	≈ 0	-336
NaI	≈ 0	1.9 ± 0.1	-291
$NaNO_3$	0.20 ± 0.06	2.73 ± 0.03	-312
$NaClO_4$	2.07 ± 0.09	4.80 ± 0.07	-246

Note that the type of thermogram and the corresponding enthalpy changes obtained in NaF are strongly reminiscent of the data typically obtained in the case of a simple 1:1 homogeneous equilibrium which has tempted some researchers to treat the data accordingly.[21] However, it is sufficient to perform DLS measurements to be convinced that the reaction product is a weakly soluble precipitate. The two successive processes (colloid complex formation and flocculation) are also apparent from the enthalpy changes recorded in $NaNO_3$(aq) and $NaClO_4$(aq). The simple homogeneous equilibrium cannot result in larger absolute values $\Delta(\Delta H)$ at higher molar ratios than at lower. As was the case in NaCl, no measurable heat effects can be observed at $r > 1$.

The formation of negative complexes (Table 3, column 3) was always slightly less enthalpically favoured (with the exception of NaF). As mentioned before, this can be explained by more pronounced repulsion of PSS chains at the negative corona that those of PAH at the surface of positive complexes. The results of calorimetric titrations of polyanion with polycation in solutions of $NaClO_4$ and $NaNO_3$ were somewhat different than in the case of other examined electrolytes. For instance, the enthalpy changes in $NaNO_3$ could be observed up to $n(\mathrm{PAH})/n(\mathrm{PSS}) \approx 1.5$ (Figure 9). The same holds for experiments performed in $NaClO_4$. It is most convenient to explain these findings by examining the data obtained at higher concentration of NaX, as described later.

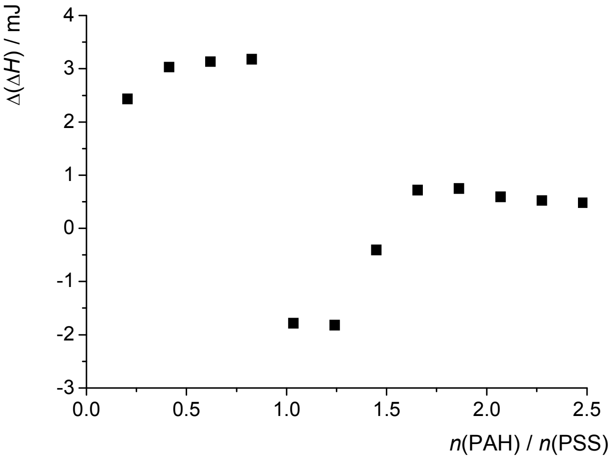

Figure 9. Successive enthalpy changes obtained by titration of polystyrenesulfonate anion ($c_\mathrm{m} = 5.0\times10^{-3}$ mol dm^{-3}, V_0 = 1.3 mL) with polyallylammonium cation ($c_\mathrm{m} = 7.5\times10^{-2}$ mol dm^{-3}) in $NaNO_3$ (c = 0.1 mol dm^{-3}), θ = (25.0 ± 0.1) ºC.

In contrast with the results of DLS measurements obtained by titration of polycation with polyanion in 0.1 mol dm^{-3} NaX, the results in 0.5 and 1.0 mol dm^{-3} (Figure 10, Figure S8, supporting information) reveal strongly specific counteranion condensation. The increase in particle size is in all cases shifted towards lower molar ratios, in particular in $NaNO_3$(aq) and $NaClO_4$(aq). Quite the opposite, the flocculation of negative complexes is only slightly affected by the increase in salt concentration, though it occurred at somewhat higher monomer ratios in $NaNO_3$ and $NaClO_4$ when compared with the other studied electrolytes.

To test whether the ion specific aggregation of positive complexes occurs, the solution of polyallylammonium chloride was titrated with sodium polystyrenesulfonate solution up to the molar ratio *n*(PSS) / *n*(PAH) = 0.5 (the solutions of reactant contained no added electrolyte). The suspensions were then titrated with 5.0 mol dm^{-3} solutions of NaX. The results (Figure 11) undoubtedly suggest that the anion specific aggregation of positive complexes occurs. The PAH chains seem to prefer the nitrate and perchlorate over the halide anions.

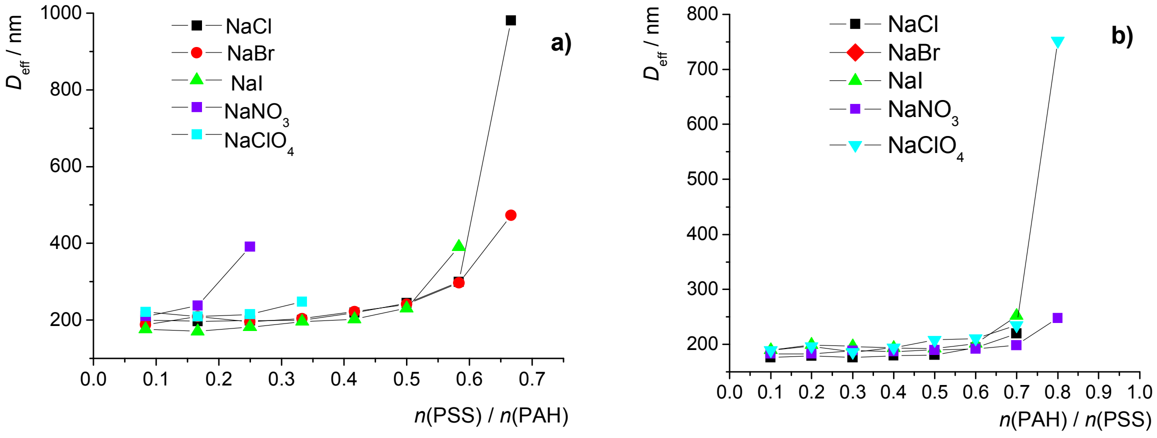

Figure 10.a) Hydrodynamic diameters of colloid complexes obtained by titration of polyallylammonium cation (c_m =1×10^{-3} mol dm^{-3}, V = 2 mL) with polystyrenesulfonate anion (c_m = 1.7 ×10^{-2} mol dm^{-3}) in NaX (aq, c = 0.5 mol dm^{-3}) at 25 ºC.

b) Hydrodynamic diameters of colloid complexes obtained by titration of polystyrenesulfonate anion (c_m =1×10^{-3} mol dm^{-3}, V = 2 mL) with polyallylammonium cation (c_m = 2.0 ×10^{-2} mol dm^{-3}) in NaX (aq, c = 0.5 mol dm^{-3}) at 25 ºC.

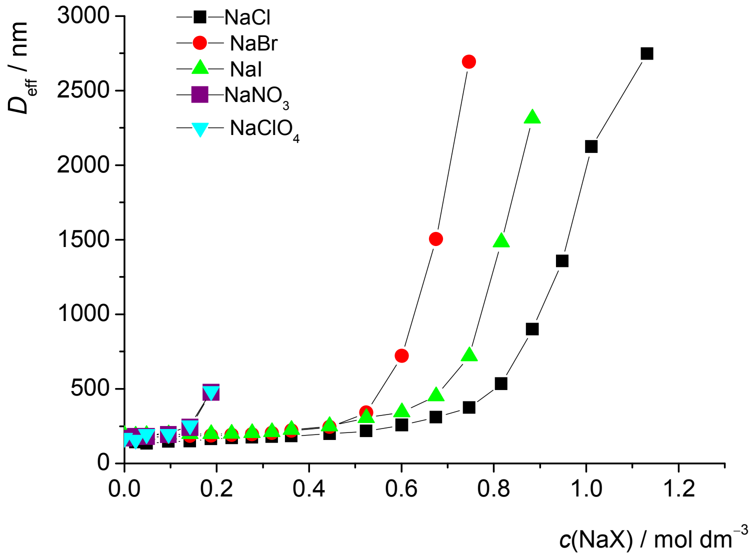

Figure 11. Hydrodynamic diameters of particles obtained by titration of positive colloid complexes (prepared by addition of sodium polystyrenesulfonate (c_m = 2 ×10^{-2} mol dm^{-3}) to solutions containing polyallylammonium chloride (c_m = 1 ×10^{-3} mol dm^{-3}, V = 2 mL); n(PSS) / n(PAH) = 0.5)) with NaX (c = 5 mol dm^{-3}) at 25 ºC.

The aggregation of positive complexes is by no means the only process occurring in solutions of $NaClO_4$ and $NaNO_3$. This is most evident from the spectra of supernatants obtained at concentrations of sodium perchlorate equal or higher than 0.5 mol dm^{-3}. The supernatant obtained by titration of polyanion with polycation at 1:1 molar ratio of monomers did not absorb in the UV region (*i.e.* the PSS was not present in supernatant), while the supernatant prepared by titration in the opposite direction (the final concentrations of reactants were the same in both experiments) contained significant amount of polyanion (Figures S9 and S10).

In contrast, the supernatants obtained in NaCl (Figure S3) and NaBr (Figure S11 and S12) showed no traces of free polyanion up to concentrations as high as 1.0 mol dm^{-3}. As noted before, spectrophotometric measurements could not be performed in concentrated $NaNO_3$ solutions due to the strong absorption of the nitrate ion. However, all other experiments suggest that, in terms of monomer pairing and precipitate composition, the situation must be very similar to $NaClO_4$.

The so far described results suggest that at higher electrolyte concentrations interpolyelectrolyte neutralisation proceeds more efficiently when the polycation is added into the solution of polyanion than *vice versa*. This is also evident from calorimetric titrations of PAH with PSS in 0.5 mol dm^{-3} $NaClO_4$ where no detectable enthalpy changes above $n(\mathrm{PSS})/n(\mathrm{PAH}) \approx 0.7$ could be observed (Figure 12 a)).Quite the opposite, the measurable enthalpy changes were recorded at molar ratios of monomers higher than 1:1 when PSS was titrated with PAH. These changes are even larger then the changes observed in 0.1 mol dm^{-3} aqueous solutions of $NaClO_4$ and $NaNO_3$ (Figure 12 b)).

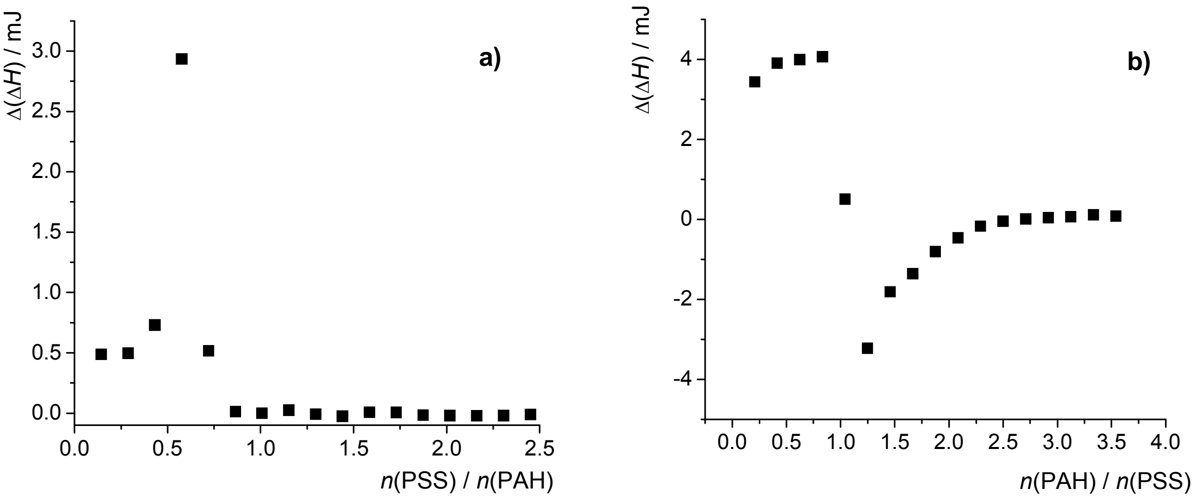

Figure 12 a). Successive enthalpy changes obtained by titration of polyallylammonium cation ($c_m = 5.0\times10^{-3}$ mol dm^{-3}, $V_0 = 1.3$ mL) with polystyrenesulfonate anion ($c_m = 6.0\times10^{-2}$ mol dm^{-3}) in $NaClO_4$ ($c = 0.5$ mol dm^{-3}), $\theta = (25.0 \pm 0.1)$ ºC.

b). Successive enthalpy changes obtained by titration of polystyrenesulfonate anion ($c_m = 5.0\times10^{-3}$ mol dm^{-3}, $V_0 = 1.3$ mL) with polyallylammonium cation ($c_m = 7.5\times10^{-2}$ mol dm^{-3}) in $NaClO_4$ ($c = 0.5$ mol dm^{-3}), $\theta = (25.0 \pm 0.1)$ ºC.

All these findings can be adequately explained by the fast (nonequilibrium) interpolyelectrolyte neutralisation and the already mentioned differences in the extent and distribution of condensed counterions. Namely, due to the greater number of directly bound counterions onto positive chains with respect to negative ones, more PAH chains can participate in non-equilibrium neutralisation of a single PSS chain than when PSS is being added into PAH. As a consequence, some of the dangling positive chains at the surface will not participate in further interpolyelectrolyte neutralisation, because of the less efficient pairing of titrand monomer units in the corona (hold and stick mechanism). In the end, less PSS monomer units will be consumed at equivalent amount of titrant added, and a significant portion of PSS chains will remain free in solution (PSS chains present in the supernatant). On the other hand, the overcharging seems to be far less pronounced when negative, primary complexes are formed (free PSS chains were not present in the supernatant at $r = 1$ for this direction even at $c(NaClO_4) = 1.0$ mol dm^{-3}. This can be explained by the more pronounced repulsion of like-charged PSS chains (consistent with the fact that the aggregation of negative complexes could be observed) when compared to positive PAH chains. Such differences between studied polyions can result in lower concentration of negative monomer units at the surface of primary negative complexes in comparison with positive ones.

The results of calorimetric titrations of PSS with PAH at molar ratios higher than 1:1 are explicable in terms of spectrophotometric, DLS and calorimetric results obtained by titration in the opposite direction. Since both aggregation and formation of reaction products containing a higher amount of positive than negative monomer units are observed when PSS is added into PAH, these processes can occur as soon as positive, secondary complexes (PAH → PSS) are formed, *i.e*, after flocculation. When this fact is taken into account, the measurable enthalpy changes at n(PAH)/ n(PSS) ≥ 1 could indicate that the incorporation of PAH into the precipitate takes place. This can lead to a notably higher amount of positive with respect to negative monomers (the enthalpy changes could be observed even at n(PAH)/ n(PSS) ≈ 2.5). The strongly asymmetric overcharging in the process of multilayer formation involving PDADMAC and PSS in concentrated NaCl(aq) was recently observed by Schlenoff and coworkers.[59] As mentioned in the introduction, the asymmetric incorporation of polyelectrolytes into the products of interpolyelectrolyte neutralisation at higher ionic strengths was first reported by Michaels[2] more than fifty years ago. The more recent investigations of Chen *et al*.[38,39] indicate that other systems behave in the similar fashion.

One should also bear in mind that the comprehensive studies of multilayers comprised of high charge density polyelectrolytes (including PAH and PSS) revealed electrolyte induced ion-specific swelling of multilayers in some cases.[40,41] This indicates that the equilibrium, as well as metastabile products of PAH-PSS neutralisation, can contain a certain amount of counterions in concentrated salt solutions. Such an effect should, for the studied polyion pair, be most pronounced in $NaNO_3$(aq) and $NaClO_4$(aq).

Perhaps the most surprising fact concerning the interpolyelectrolyte neutralisation of PAH and PSS at higher electrolyte concentrations, especially in $NaNO_3$(aq) and $NaClO_4$(aq), is that the process remains approximately isoenthalpic irrespective of electrolyte type (Table 4).

Table 4. Complexation thermodynamics of polystyrenesulfonate anion with polyallylammonium cation; enthalpies of formation of charged colloid complexes in NaX (c = 0,5 mol dm^{-3}); θ = (25.0 ± 0.1) ºC.

NaX	$(\Delta_c H_{\mathrm{PSS}\rightarrow\mathrm{PAH}} \pm \mathrm{SE})$/ kJ mol^{-1}	$(\Delta_c H_{\mathrm{PAH}\rightarrow\mathrm{PSS}} \pm \mathrm{SE})$/ kJ mol^{-1}
NaF	[a]	[a]
NaBr	-1.37 ± 0.07	≈ 0
NaI	0.40 ± 0.07	1.7 ± 0.2

$NaNO_3$	[b]	1.44 ± 0.05
$NaClO_4$	[b]	3.77 ± 0.07

[a] The calorimetric experiments were not performed in NaF(aq) at $c \geq 0.5$ mol dm^{-3}.

[b] Enthalpies of formation of charged colloid complexes could not be determined due to formation of precipitate which contained large excess of PAH monomers.

The formation of reaction products containing higher amount of positive with respect to negative monomer units is therefore difficult to explain in terms of enthalpic (counterion condensation to unpaired, excess PAH monomers induced by their incorporation in precipitate up to neutrality) and entropic (the benefit of counterion release upon monomer pairing) balance. The interpolyelectrolyte neutralisation would otherwise have to be more enthalpically unfavourable than at lower concentrations of electrolyte. The reason for higher amount of PAH than of PSS monomer units incorporated in precipitate for the herein investigated polyion pair could simply be the notable screening of polycation charge with counteranions at higher ionic strengths. In other words, the unpaired functional groups of PAH in precipitates and complexes, in terms of counterion condensation, seem to be quite similar to free PAH monomer units in solution.

As stressed on several occasions, the equilibrium is reached extremely slowly. Even after eight weeks under constant mixing at room temperature ($\theta \approx 25$ °C) the spectra of supernatants obtained by titrations in both directions at $r = 1$ and $c(NaClO_4) = 1.0$ mol dm^{-3} differed (Figure 11 a)). In contrast, relatively rapid establishment of equilibrium at elevated temperatures ($\theta \approx 60$ °C) under the same ionic conditions was observed. The equilibrium product of the reaction at 1:1 molar ratio of oppositely charged monomer units was a very weakly soluble polysalt containing an equal amount of positive and negative functional groups (*i.e.* the supernatant showed no evidence of free polycation, Figure 13).

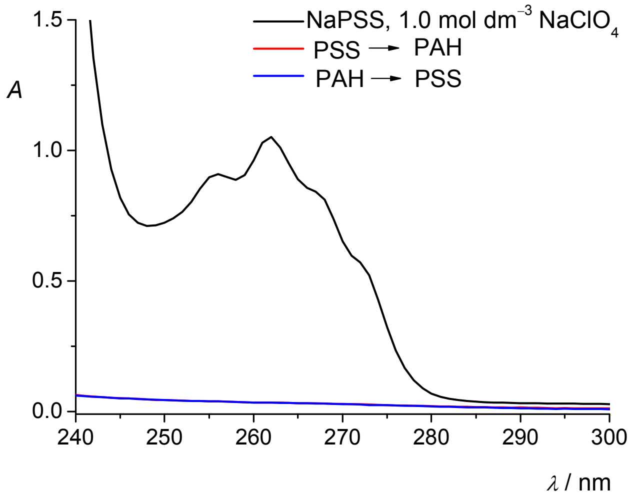

Figure 13. Spectra of supernatants obtained by centrifugation of suspensions prepared by titration of polycation with polyanion (PSS→PAH) and *vice versa* (PAH→PSS) at $r = 1$ and left to equilibrate for a week at $\theta \approx 60$ °C . (r – ratio of oppositely charged monomers. NaPSS - spectrum of sodium polystyrenesulfonate ($c_{\mathrm{m}} = 3.3 \times 10^{-3}$ mol dm^{-3}), in NaClO$_4$ ($c = 1.0$ mol dm^{-3}).

In principle, such a finding does not necessarily mean that the reaction is quantitative at 25 °C. Upon cooling the reaction mixture from 60 °C to 25 °C the equilibrium establishment (polysalt dissociation) can again be slow, so the spectra recorded at 25 °C might actually reflect the equilibrium composition at higher temperature. Relative permittivity of water decreases with temperature,[60] thus favouring counterion condensation. On the contrary, the anion hydration becomes slightly more favourable as water temperature increases.[61] Weak temperature dependence of polyelectrolyte osmotic coefficients[62] indicates that these two opposing effects largely cancel each other. Consequently, the process should be quite favourable in terms of entropy even at high temperatures. This is confirmed by calorimetric experiment performed by adding PAH to PSS solution at 60 °C and c(NaClO$_4$) = 1 mol dm^{-3} (Figure 14). The interpolylectrolyte neutralisation at this temperature is virtuallly isoenthalpic (slightly endothermic), but still quantitative (Figure 14), at least after the equilibrium had been reached. This is strong evidence that the equilibrium product obtained at the same ionic conditions and in solutions of all investigated electrolytes at 25 °C should be very weakly soluble polysalt. Namely, the complexation process at this temperature is enthalpically most demanding in NaClO$_4$(aq).

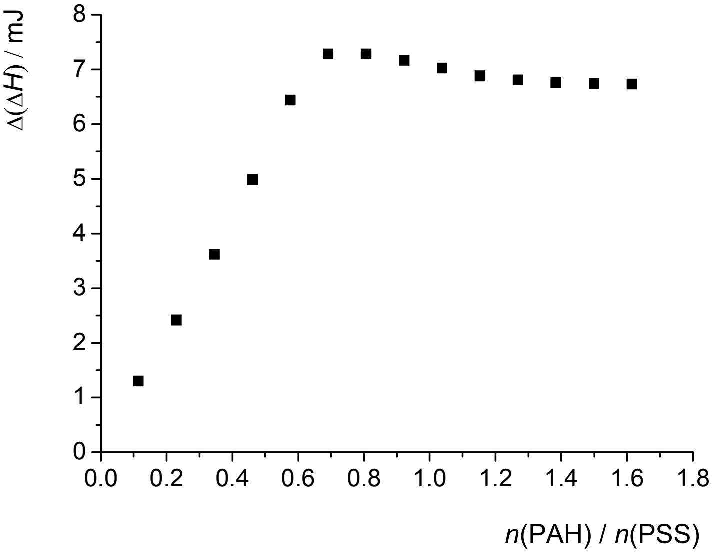

Figure 14. Cumulative enthalpy changes obtained by titration of sodium polystyrenesulfonate anion ($c_m = 5\times10^{-3}$ mol dm^{-3}, $V_0 = 1.3$ mL) with polyallylammonium cation ($c_m = 5\times10^{-2}$ mol dm^{-3}) in NaClO$_4$ ($c = 1.0$ mol dm^{-3}), $\theta = (60.0 \pm 0.1)$ ºC.

Apart from the described results, the already mentioned quantitative isoenthalpic, interpolyelectrolyte neutralisation of PDADMAC and PSS (chapter 3.1)[18] at quite high NaCl concentration and room temperature is jet another indication that the isoenthalpic neutralisation of high charge density polyelectrolytes should be quantitative even at quite high concentration of simple electrolyte. As stated before, the entropy increase caused by counterion release from highly-charged polyions seems to be strongly favourable. However, the equilibrium establishment at room temperature can be extremely slow.

As noted in the introduction, the strongly anion-specific built-up of PDADMAC-PSS multilayers was observed in the case when simple 1:1 electrolyte was added to the solution of reactants.[42-44] The results suggest that such an effect is expected for the corresponding PAH-PSS multilayers. The investigations indicate that the amount of deposited material in PAH-PSS PEMs should be the largest in the case of NaNO$_3$ and NaClO$_4$. As noted before, the reason for expected (PEMs) and observed (interpolyelectrolyte neutralisation of PAH and PSS in solution) anion-specific influence does not lie in the large difference in the energetics of interpolyelectrolyte in the presence of different anions ($\Delta_{\mathrm{c}}H$ are rather similar irrespectively of electrolyte type), but in differences in anion distributions around PAH. This might also be the case for the PDADMAC-PSS system. According to the results concerning the composition of metastable reaction products in solution, the observed increase in PAH-PSS multilayer thickness[32,33] upon the increase in ionic strength should be the consequence of more pronounced, asymmetric overcharging and the increase in extrinsically (counteranion) compensated functional groups (especially in the case of polycation). In contrary to expectations, the calorimetric measurements indicate that these effects cannot be explained by the enthalpically more demanding neutralisation at higher electrolyte concentrations (minute dependence of complexation enthalpy on electrolyte concentration), but with fast, non-equilibrium neutralisation and the changes in the distribution of counterions upon the increase of ionic strength (double layer contraction).

Another factor with potentially large influence on structure and composition of polyelectrolyte based nanostructures still remains to be explored, and that is the effect of the solvent. Since some solvents solvate ions much more strongly than water, considerable favourable influence on the binding energetics can be expected. On the other hand, the solvents of poorer quality are expected to make the process more enthalpically unfavourable, but more favourable in terms of entropy related with counterion release. However, many interesting phenomena regarding reaction kinetics and asymmetric neutralisation can occur. Additionally, polymer collapse or highly reduced solubility of reactants can also be expected.[63]

4. Conclusion

On the basis of described results several important conclusion regarding specific and general aspects of counterion condensation onto high charge density and high P_n polyions can be deduced. First of all, the chains of two investigated polyions are notable different in terms of counterion condensation. The PAH double-layer seems to be more contracted than that of PSS. This is most likely due to differences of the charge distribution with respect to the chain backbone. Namely, the positively charged amino groups of PAH are situated much closer in electrostatically favoured *entgegen* configuration with respect to vinyl units than are the monomer groups of PSS. As a consequence, PAH chains require more directly bound counterions than the chains of PSS. This then leads to large differences in composition and stability of primary positively and negatively charged complexes, as well as to formation of precipitates containing larger amounts of PAH monomer units with respect to PSS charged groups. The described differences in the counterion distributions around PAH and PSS can serve as a clear remainder of how limited the structureless models of highly charged polyions[64–66] are, despite the fact that they successfully capture their essential physico-chemical properties in solutions.

The second conclusion concerns the origin of ion-specific effect. When the reaction between PAH and PSS is studied in the low-electrolyte regime there is almost no influence of anion type on the product composition and, importantly, on the energetics of interpolyelectrolyte neutralisation. On the other hand, the increase in salt content reveals quite specific ionic distributions around PAH. Clearly, the direct binding of weakly hydrated perchlorate and nitrate ions is favoured over the direct binding of halogen anions. The reason why they can not be observed at lower ionic strengths must hence lie in the fact that the overall number of condensed counterions (in the chain vicinity and at larger distances) remains low. That is, the chains are notably extended and the double-layers spread far into the solution. In terms of solvation, direct binding will be favoured with ions of low charge density and indirect with those of high charge density. Still, the unfavourable repulsions along the chain have to be reduced so a larger number of indirectly bound ions will be needed with respect to directly bound. Macroscopically, the two opposing effect seem to largely cancel, since osmotic coefficients[67–69] and apparent dissociation constants of polyacids[52,70,71] in general remain similar irrespectively of counterion type (the overall number of immobilized ions must hence be similar). The increase in ionic strength results with notable double layer contraction and more pronounced ion-pair type of binding (direct, solvent shared and solvent separated pairs). This leads to notable different anion distributions in the polyion vicinity (evident from the results of DLS measurements of primary complex size in solutions containing different anions).

Interestingly, the interpolyelectrolyte neutralisation of studied polyion pair remained isoenthalpic irrespectively of the counterion type and concentrations. It seems that, despite the mentioned differences in the counterion distributions, the overall energetics involving the ionic atmosphere (consisting of directly and indirectly bound cations) removal must be similar. This could also hold for other systems containing highly charged particles and polymers where the electrostatic effect dominate (*i.e.* both PAH and PSS monomers do not bind the ions of examined electrolytes except at quite high ionic strengths). Perhaps the reasons for many ion-specific effects in colloid and surface chemistry also do not lay in large differences in energetics of counterion binding (direct and indirect), but in differences in counterion distributions in the vicinity of charged groups. This could explain why many ion specific effects follow Hofmeister series,[66] largely consistent with the standard hydration parameters of ions.[57] Namely, the amount of directly bound counterions will be largely determined by their hydration and the related charge density. This has been shown to be the case for several other highly charged systems by Cremer *et al.*[72] recently. Of course, other effect should be considered as well, especially with molecular ions comprised of several atoms capable of hydrogen bonding and multidentate binding (ammonium, perchlorate, sulphate, nitrate, *etc.*).[66] A considerable lack of calorimetric data for the time being prevents further conclusions to be drawn.

The third quite important conclusion concerns the equilibrium composition and the equilibrium position in systems containing high charge density and high P_n polyelectrolytes. It should be clearly stated that the apparent reversibility of interpolyelectrolyte neutralisation (similar composition of reaction product irrespectively of the addition order)) must no longer be considered as evidence that chemical equilibrium has been reached. The results of present investigation clearly indicate that the presence of electrolyte is prerogative for equilibrium which is accomplished extremely slowly even at elevated temperatures. However, the increase in electrolyte concentration at first disrupts the monomer pairing thereby favouring the formation of asymmetric (containing higher amount of one monomer with respect to the other), metastable products. These findings indicate that similar behaviour can occur in all systems in which phase separation occurs at lower monomer ratios. Namely, the aggregation of primary complexes may not be the only effect taking place at lower monomer ratios. Surprisingly, the formation of asymmetric metastable products was found to be isoenthalpic as well, which indicates that the chains of the polyion present in excess must, in terms of counterion condensation, be quite similar to free chains in solution.

One other import finding is that the formation of positive or negative complexes is enthalpically quite similar to formation of flocculate containing mostly paired monomers. This fact, overlooked in several previous investigations,[21,22] is the strong evidence that the notable overcharging of PECs and PEMs is largely due to dangling chains protruding from the surface of the mostly neutral core. In contrast with largely held view, the increase in electrolyte concentration affected the enthalpies of interpolyelectrolyte neutralisation weakly. This finding somewhat surprisingly suggests that the notable double layer contraction with increase in ionic strength occurs with little changes in enthalpy of counterion condensation

Apart form resolving some of the fundamental aspects of interpolyelectrolyte neutralisation in solution and at surfaces, the presented results can also be of practical purpose. The displayed long-term metastability of PAH/PSS nanocomposites makes them perfectly safe for the long term use in biomedicine and biotechnology.[49–51] Additionally, the observed strong influence of counterion type on the composition of PAH/PSS nanoassemblies provides a path for the modification of their properties simply be changing the type of salt during their preparation. In particular, the strongly asymmetric composites containing higher amounts of PAH than of PSS can be prepared in concentrated solutions of sodium perchlorate and nitrate. This result could be used in cell culture, since the cells adhere to positively charged surfaces.[49,51] Of course, the anions would have to be exchanged with chlorides (physiological solution). The long-term stability of PAH/PSS multilayers indicates that such post-modification is plausible. It is also known that the mechanical properties (elastic modulus) of PEM and PEM nanotubes depend on the ionic conditions used for their preparation.[73 ,74] Perhaps, the mechanical properties of PAH/PSS nanoassemblies could be adjusted by choosing the appropriate concentration and type of added salt. The ions counterbalancing the charge at the surface of PEMs also enable significant modification of surface wettability.[75] The results on PAH/PSS system reported in this paper could be used for better tuning of this important property as well.

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