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ORIGINAL CONTRIBUTION

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Salt influence in the polymer–surfactant interaction in solution. A fluorescence probe investigation of the EHEC/SDS/water system

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Abstract The effect of small amounts of salt on the interaction between two fractions of ethyl(hydroxy)ethyl cellulose, EHEC, and sodium dodecyl sulfate, SDS, has been investigated by means of steady-state fluorescence measurements. The two polymer fractions display different properties in hydrophobicity expressed as different cloud points. The results are discussed in relation to hydrodynamic (viscosity) and thermodynamic (equilibrium dialysis) properties. The micropolarity as sensed by the probe pyrene shows that the polymers begin to interact with SDS at a lower concentration in the presence of salt. The average aggregation numbers of polymer-bound clusters, N_p, were obtained by fluorescence-quenching data in combination with equilibrium dialysis experiments. N_p was found to increase in the presence of salt for the EHEC fraction with a high cloud point (CP). The polymer with a low CP displays higher N_p in the presence of salt at low SDS concentrations, but exhibit lower N_p at higher SDS concentrations than in the salt-free system. The microviscosity index as determined by intramolecular excimer formation of 1,3-di(1-pyrenyl)propane (P3P) is highest for the lowest N_p and there is a correlation with N_p in the presence as well as absence of salt for both EHEC fractions. It is found that when the same fractional amount of SDS is bound to the polymers, 10–20% of the value of saturation, the increase in macroviscosity occurs and the microviscosity shows high rigidity.

Key words Ethyl hydroxyethyl cellulose – sodium dodecyl sulfate – interaction – sodium chloride – microviscosity – average aggregation number – micropolarity

Introduction

Several water-soluble uncharged polymers interact with anionic surfactants in solution and thereby give the system special rheological properties [1–5]. This phenomena offers an interesting feature for pharmaceutical formulations, since many drug molecules have an amphiphile character and charged polymers are commonly used in preparations as thickeners and flow modifiers. Over the last decade this interaction pattern has been thoroughly investigated and several reviews exist [6–8]. Previous publications from this laboratory [9–15] have mainly discussed the properties of the EHEC/SDS/water system in the absence of salt, but a strong sensibility to added electrolytes has also been reported [16]. It is of importance to investigate the influence of salt in the perspective of polymer/surfactant systems as pharmaceutical excipients, since salt is present in all body fluids. The effect of salt on the phase conditions of the EHEC/SDS/water system at high polymer concentrations has been studied by Carlsson et al. [17, 18], and altered interactions between a surfactant and an uncharged polymer in the presence of salt has been reported by others [1, 19–21]. To provide an insight of the influence of salt on the interaction pattern between EHEC and SDS at low polymer concentrations, a fluorescence probe study has been performed, in addition to the previous investigation concerning the hydrodynamic and thermodynamic effects of salt of this particular system [16].

Over the last two decades, fluorescence spectroscopy has proven to be a powerful tool for obtaining information on the microstructure of aqueous polymer/surfactant systems [22–24]. In this study, we use fluorescence probe techniques to yield average aggregation numbers of polymer-bound surfactant clusters as well as information on micropolarity and microviscosity in the absence and presence of NaCl. Contrary to bulk viscosity, which depends on the resistance of the solution to flow, the microviscosity, as sensed by a hydrophobic fluorescent probe, gives information about the probe motion on a molecular level, such as within the micelle or a polymer-bound cluster. This is of importance since many drug molecules are of hydrophobic character, and when solubilized in clusters, the microenvironment may affect the release rate of the drug.

Like many other polysaccharides, EHEC displays a complex behavior, due to its mixed hydrophobic/hydrophilic structure, when added to a surfactant solution. This quality of EHEC also results in pronounced effects in water solutions of the polymer [25], for instance, the coil–coil interaction increases with temperature, leading to phase separation at a specific temperature; cloud point (CP). This temperature-dependent interaction is further promoted by addition of surfactants and salt. The extent to which the macromolecule is affected by additives is partly determined by the balance between hydrophilic and hydrophobic effects in the binary case. Thus, two fractions of EHEC were used in this study displaying different cloud points, 30 and 65 °C, respectively for 0.20% EHEC/water solutions. Both EHEC fractions interact with SDS in solution mainly in a similar way, although there are important differences and observed effects are more pronounced for the polymer with a low CP [16]. Results obtained from steady-state fluorescence measurements in terms of micropolarity, average aggregation number and microviscosity correlates well with the macroscopic properties of both systems. When small amounts of salt is added to both systems, the properties of the respective polymer/surfactant solutions differ from each other, and that the effects are more pronounced for E230G, the fraction with a high CP. The obvious effects of salt are found at low surfactant concentrations, probably, since at these compositions the contribution of 5 mM NaCl to the ion strength in high and diminish upon raising the SDS concentration. Also, in the presence of large amounts of surfactant the polymer is found to be more solubilized and thus, better sheilded from the added salt. The results are discussed in connection with the other essential characteristics of the EHEC/SDS/water system.

Experimental section

Materials

The following chemicals were used as supplied: sodium dodecyl sulfate (SDS), 99.9% pure, MERCK, Darmstadt, Germany. Radioactive SDS, ^{35}S, Amersham, Buckinghamshire, England. Sodium chloride, 99.5% pure, MERCK, Darmstadt, Germany, 1, 3-di(1-pyrenyl)propane (P3P) from Molecular Probes, Eugene, OR, U.S.A., tris(2, 2′-bipyridyl)ruthenium(II)chloride ($Ru(bipy)_3^{2+}$) and 9-methylanthracene (9-MA) (98%) from Aldrich-Chemie, Steinham, Germany. Pyrene, (98 + %) from Acros Chimica, Belgium, was twice recrystallized from absolute ethanol. Ethyl(hydroxyethyl)cellulose (EHEC) fraction CST-103 $M_n = 480\,000$, $MS_{eo} = 0.7$, $DS_{ethyl} = 1.5$, L920 32, and fraction E230G, $M_n = 5\,000\,000$, $MS_{eo} = 0.9$, $DS_{ethyl} = 0.9$, L92003, were supplied by Akzo Nobel, Stenungsund, Sweden. The fraction called CST-103 will hereafter be referred to as CST. The EHEC stock solution was freed from remaining salt by dialysis in tube membranes (MW cut-off approximately 10 000) from Union Carbide, Chicago IL. The stock solution of EHEC was filtered through 0.8 μm Millex-AA filters, Millipore SA, Molsheim, France. All solutions were made with MilliQ water (Millipore) as solvent.

Preparation of solutions

Stock solutions

Stock solutions of the EHEC fractions were prepared according to a standard technique described elsewhere [9]. The stock solutions of SDS and NaCl were prepared by dissolving the substances in water in appropriate concentrations. Radioactive SDS was dissolved in water and diluted to obtain adequate activity for dialysis experiments.

Fluorescence measurements

For experiments with P3P, the probe was dissolved in acetone, *pro analysi* (Merck, Darmstadt, Germany), and aliquots of the probe-acetone solutions were added to the EHEC/SDS/water solutions to achieve a final approximate probe concentration of 1×10^{-6} M P3P. In order to avoid formation of P3P-microcrystals, a problem also described with the closely related probe dipyme [26], the P3P-aliquots were added to a 100 mM SDS stock solution and then further diluted. The final EHEC/SDS/water/P3P-solutions were kept on a rotating table in the dark for at least three days until equilibrated. For quenching experiments with $\mathrm{Ru(bipy)_3^{2+}}$-9-MA as the probe-quencher pair, the EHEC/SDS/water solutions contained approximately 1×10^{-6} M $\mathrm{Ru(bipy)_3^{2+}}$ and between 0.1 and 5×10^{-4} M 9-MA. The quencher was incorporated into a small amount (5 ml) of each EHEC/SDS/water-probe solution by dissolving approximately 5 μl of an acetone-quencher solution. These solutions were then mixed with the corresponding quencher-free solutions to achieve a set of appropriate concentrations with respect to 9-MA.

Viscosity

Appropriate amounts of the EHEC stock solution were added to diluted surfactant solutions. In the solutions containing salt, small portion of NaCl stock solution was added to the SDS solutions before the polymer was added. All solutions measured at room temperature were prepared 24 h in advance to let the time-dependent processes settle [10].

Equilibrium dialysis

A complete dialysis study of a certain polymer–surfactant system comprised a series of solutions, i.e. a polymer solution of a specific concentration and a set of surfactant solutions. Small amounts of radioactive SDS were added to the surfactant. In the experiments with salt present, the SDS stock solution was added to an appropriately diluted NaCl stock solution. Sodium chloride was also added to the EHEC solutions so that concentration equalled that of the surfactant solution.

Methods

All experiments, with both EHEC fractions, have been performed with a constant polymer concentration of 0.20% (w/w), varying the SDS content from 0 to 20 mM and the amount of added salt between 0 and 5 mM at 20 °C. The polymer concentration was chosen as to be below the critical overlap concentration, c^* [9], (the concentration-point of entanglement) but still high enough to give marked results. The experimental data, from various techniques, showed that the effects of added salt in the EHEC/SDS/water system increase gradually with NaCl concentration, thus, in the figures only the results for 0 and 5 mM of salt are presented with the understanding that the graphs corresponding to 1 and 3 mM NaCl are in between those shown. In a previous work CP measurements were made [16], which have been used in this study to ensure that phase separation was not present in the solutions. An important feature of polymer–surfactant systems is the surfactant concentration corresponding to the onset of interaction. In the literature [8], this concentration is commonly named as the critical aggregation concentration (CAC) defined as the critical micelle concentration (CMC) in the presence of polymer. Although the EHEC/SDS/water system does not show a clear-cut cooperative mechanism, we have chosen to denote the foot point in the absorption isotherm as CAC in order to simplify the discussion.

Fluorescence

Steady-state fluorescence experiments were used to study the CMC for pure surfactant both in the absence and presence of salt, as well as the CAC, for the EHEC/SDS/NaCl/water system by determining the hydrophobic index, I_1/I_3, for the systems. It is well established [27] that the ratio of the first and third vibrational peak in the emission spectrum of pyrene, I_1/I_3, is a measure of the micropolarity it senses in a certain environment. The hydrophobic probe pyrene dissolved in pure water gives a I_1/I_3-value of approximately 1.9 and I_1/I_3 decreases when the polarity of the medium decreases. It has been shown that a sudden drop in I_1/I_3 occurs when micelles are formed at CMC for binary surfactant/water systems [27] and at CAC in the presence of polymer [11, 12, 28–30].

The values of average aggregation numbers of polymer-bound clusters, N_p, have been obtained using the steady-state fluorescence-quenching (SSFQ) method developed by Turro and Yekta [31]. In recent publications from this laboratory, SSFQ has been utilized [11, 12, 30] and validated [12] on non-ionic cellulose ether/SDS/water systems. N_p was obtained by combining fluorescence-quenching data with results from equilibrium dialysis as described earlier [11]. When $[\mathrm{SDS}]_{\mathrm{eq}}$ exceeds the CMC, "normal" free micelles are present in the EHEC/SDS/water solution as well. For these compositions N_p has been calculated with a correction for the normal micelles using the CMC-values extracted from Fig. 1 and average aggregation numbers of free micelles in binary SDS/water

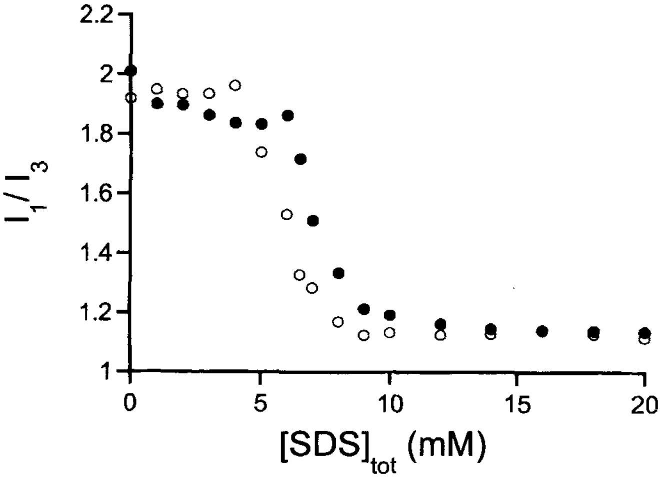


Fig. 1 The hydrophobic index of pyrene, I_1/I_3, as a function of the total SDS concentration, $[SDS]_{tot}$, at 20 °C in binary SDS/water solutions. (●) no salt added; (○) 5 mM NaCl

solutions [11]. The average aggregation numbers for normal micelles were found to be in the order of 60–70 for the NaCl concentration range 0–5 mM which is in good agreement with the literature [31–34].

The microviscosity of the EHEC/SDS/NaCl/water systems has been explored utilizing the probe 1,3-di(1-pyrenyl)propane, P3P, which first was used by Zachariasse in 1978 [35] on the binary system SDS-water, and found to agree well with fluorescence depolarization measurements. P3P and the closely related probe 1,1-dipyrenyl methyl ether, dipyme [36], have been used in the literature to explore the microviscosity of aqueous surfactant systems [35, 37–39] as well as amphiphilic self-aggregating polymers [26, 40, 41]. In a recent work from this laboratory [15], P3P was found to be suitable for microviscosity-studies on the system EHEC/SDS/water and the results correlate well qualitatively with steady-state depolarisation results using perylene as well as with results from intramolecular relaxation about bonds of *p*-(*N*-dimethylaminobenzylidene), BMN. P3P has the ability to form intra-molecular excimers allowing it to be used at very low concentrations thereby avoiding the high concentration errors of intermolecular excimer-forming probes i.e. pyrene [42]. The extent of intra-molecular excimer formation and emission of P3P is dependent on the local friction of the probe imposed by its microenvironment. Hence, the monomer to excimer intensity ratio, I_M/I_E, provides a qualitative index of the microviscosity as sensed by the probe. There are several difficulties in extracting microviscosity-information from fluorescent probe techniques [23, 42–44] such as the location of a probe in and the perturbation of a cluster or micelle, as well as the use of an isotropic reference system with known viscosities as a calibration curve. For this reason, the values herein represent relative changes within the microenvironment and there is no attempt to present absolute values of microviscosity.

All fluorescence measurements were recorded on a SPEX Fluorolog 2 model FL1T2 steady-state spectrofluorometer in the "s"-mode with 0.5 mm excitation- and emission slits. The quvette holder was thermostated to 20 °C using a circulating water bath. Pyrene was excitated at $\lambda = 334$ nm and emission spectra were recorded. The I_1/I_3-ratio was taken as the first ($\lambda \approx 377$ nm) and third ($\lambda \approx 388$ nm) peak-height-ratio in the fine vibrational spectrum of pyrene. Steady-state fluorescence quenching experiments were performed with $Ru(bipy)_3^{2+}$ as the probe and 9-MA as the quencher. $Ru(bipy)_3^{2+}$ was excitated at $\lambda = 450$ nm and the emission was recorded at $\lambda = 625$ nm. In these experiments, the emission slit was set to 1.25 mm. SSFQ works well for determination of aggregation numbers if certain criteria are met [45, 46], including the use of a highly efficient probe-quencher pair with very low fraction in the aqueous phase, and that the cluster-sizes determined do not exceed about 100. P3P was excitated at $\lambda = 348$ nm and emission spectra were recorded between 350 and 500 nm. The I_M/I_E ratio was taken as the monomer ($\lambda \approx 377$ nm) to excimer ($\lambda \approx 485$ nm) peak-height-ratio. All fluorescence measurements were run in duplicates and the error was 5% or less.

Viscosity

The reduced viscosity, η_{red}, for each EHEC/SDS solution was calculated according to standard procedures [47]. The "solvent viscosity" used were taken as that of the corresponding SDS/water and SDS/NaCl/water solution, respectively. The viscometric measurements were performed in ordinary capillary viscometers with a flow time for water of approximately 100 s at 20 °C. The samples were thermostated in the viscometer 15 min before the measurements were made. Corrections for kinetic and end effects were found unnecessary [47].

Equilibrium dialysis

SDS is bound to EHEC in the form of clusters or mixed micelles [11], and when the adsorption isotherms, as seen in Figs. 6a and b, were calculated for the salt-free systems it was assumed that the bound ions make no contribution to the ionic activities, i.e. the degree of dissociation, α, is 1. Thus, the effect of the Donnan equilibrium is given by standard equations [14] where $[SDS]_{eq}$ is the surfactant concentration in equilibrium with the SDS-EHEC aggregates. The dialysis equilibrium can be expressed as

$$[\mathrm{SDS}]_{\mathrm{tot}} = [\mathrm{SDS}]_{\mathrm{eq}} + c_p y \,, \qquad (1)$$

where c_p is the polymer concentration represented in grams per litre, and y is expressed as millimoles of SDS adsorbed per gram of polymer. The Donnan effect diminish to some extent when small amounts of salt are present in the dialysis solutions, but it still needs to be considered [14]. This was done according to previously described procedures [16]. To avoid the Donnan effect completely the sodium chloride concentration should be not less than 0.1 mol [48, 49]. The equilibrium dialysis experiments were carried out in specially designed cells, with retentate and dialysate compartments separated by a SPECTRA/POR membrane (MW cut-off 12–14000). The cell design was similar to the one used by Fishman and Eirich [48]. Each series of experiments was carried out with polymer solutions of constant concentration placed in the retentate compartments but with surfactant solutions of various concentrations placed into the respective dialysate compartments as performed earlier [14]. After the cells had reached equilibrium, the amount of SDS was determined and the adsorption isotherm could be calculated [9]. Both the polymer solution and the surfactant solutions contained equal NaCl concentrations in each experimental series (salt present).

Results

Fluorescence measurements

The variation of I_1/I_3 versus the total SDS concentration, in the absence of polymer, is presented in Fig. 1. It can be seen that the normal CMC for the surfactant is lowered upon addition of salt, as has been reported by others [20, 50]. Figures 2a and b show the variation of I_1/I_3 with the total SDS concentration for EHEC fractions CST-103 and E230 G, respectively. When 5 mM NaCl is added to the systems, the drops in I_1/I_3 occur at somewhat lower SDS concentration for both EHEC fractions, indicating a decrease in CAC with respect to the SDS concentration. After the drop in I_1/I_3, its value levels off to an asymptote which for the quarternary SDS/EHEC/NaCl/water systems is lower than in the case of the ternary SDS/EHEC/water systems. The E230/SDS/water system, with and without NaCl, display similar features compared to the CST/SDS/water system with the exception that the CAC occurs at higher SDS concentrations.

In Figs. 3a and b, the average aggregation number of polymer-bound SDS clusters, N_p, is presented as a function of $[\mathrm{SDS}]_{\mathrm{tot}}$ for the EHEC systems both in the presence and absence of salt. In the absence of salt the clusters start to form on CST at the CAC, after which there is a considerable increase in N_p with $[\mathrm{SDS}]_{\mathrm{tot}}$. N_p reaches a maximum at about 15 mM SDS, where after a slight decrease in N_p is seen when the SDS concentration approaches 20 mM SDS. This decrease may be related to the decline of the dialysis curve. When salt is added to the CST/SDS/water system, N_p is altered. The average aggregation number shows a somewhat higher value at low $[\mathrm{SDS}]_{\mathrm{tot}}$ compared to salt-free conditions, although at a total SDS concentration of approximately 12 mM, the lines cross and N_p now displays a lower value in the presence of salt. The E230G/SDS/water system, with no salt present, displays similar features as the CST/SDS/

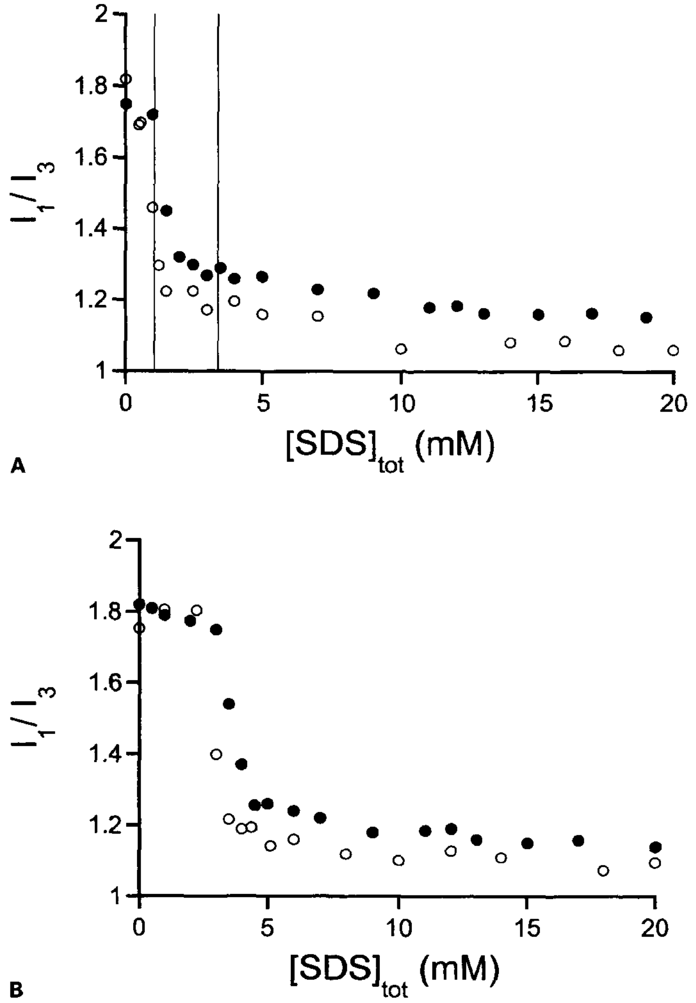


Fig. 2 The hydrophobic index of pyrene, I_1/I_3, as a function of the total SDS concentration, $[\mathrm{SDS}]_{\mathrm{tot}}$, at 20 °C in 0.20% EHEC/water solutions. (●) no salt added; (○) 5 mM NaCl. (a) EHEC fraction-CST-103, (b) EHEC-fraction E230 G. Area between vertical lines in (a) indicates phase separation in the presence of 5 mM NaCl

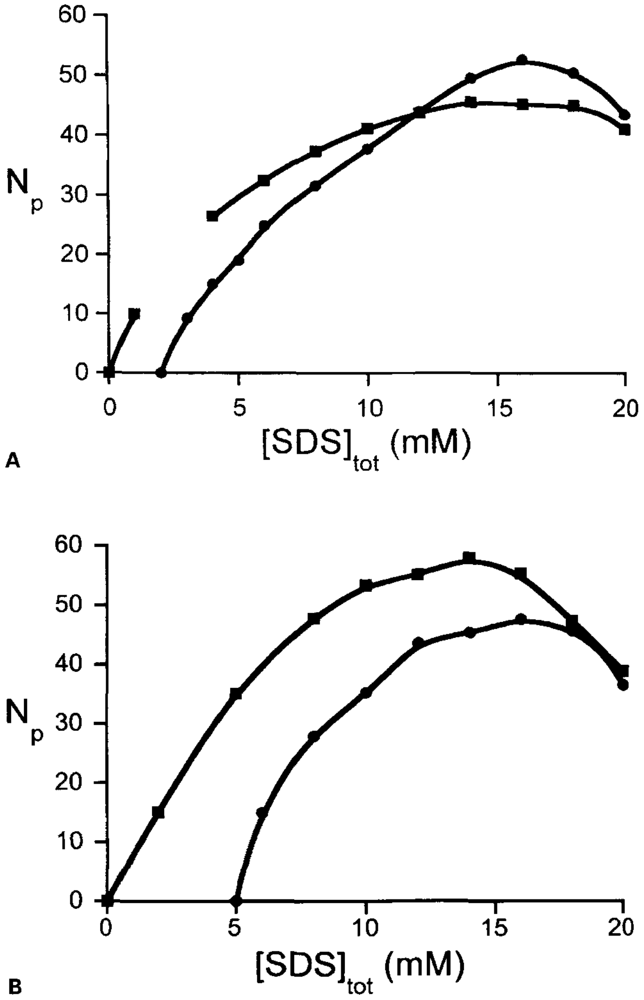


Fig. 3 Average aggregation numbers of polymer-bound SDS clusters, N_p, formed in 0.20% EHEC/water solutions as a function of the total SDS concentration, $[SDS]_{tot}$, at 20 °C. (●) no salt added; (■) 5 mM NaCl. (a) EHEC-fraction CST-103 (b) EHEC-fraction E230 G. Area not connected by the curve in (a) indicates phase separation in the presence of 5 mM NaCl

water system, with the exception that the clusters start to form at higher $[SDS]_{tot}$ and the maximum in N_p is not as high. In presence of NaCl, SDS begins to form clusters on E230G at a lower $[SDS]_{tot}$. N_p is also higher, but the difference in aggregation numbers diminish with increasing SDS concentration to finally vanish at $[SDS]_{tot} = 16$ mM.

Figures 4a and b show the microviscosity index, I_M/I_E, as a function of the total SDS concentration for EHEC fractions CST and E230 G, respectively. For both polymers I_M/I_E increases abruptly at CAC followed by a maximum, and with increasing concentration of SDS, after this maximum, I_M/I_E decreases. The CST curve in absence of salt show a weakly developed "shoulder" at $[SDS]_{tot} = 7$ mM. For the binary system SDS/water, an increase in I_M/I_E is seen at the CMC up to about $I_M/I_E \approx 1$ and I_M/I_E remains between 1 and 2 in the SDS-concentration-range 0–100 mM [15]. The EHEC/SDS/water systems under present study show higher indices of microviscosity for all compositions shown as compared to the binary system SDS/water with respect to the SDS concentration. When 5 mM of NaCl is added to the CST/SDS/water solution, the abrupt increase in I_M/I_E occur at a lower SDS concentration, whereafter a region of phase separation is present, as seen in Fig. 4a. After the phase separation, at 4 mM of SDS, there is a decrease in I_M/I_E, although its slope is less steep compared to the salt-free case. When salt is added to a E230G/SDS/water

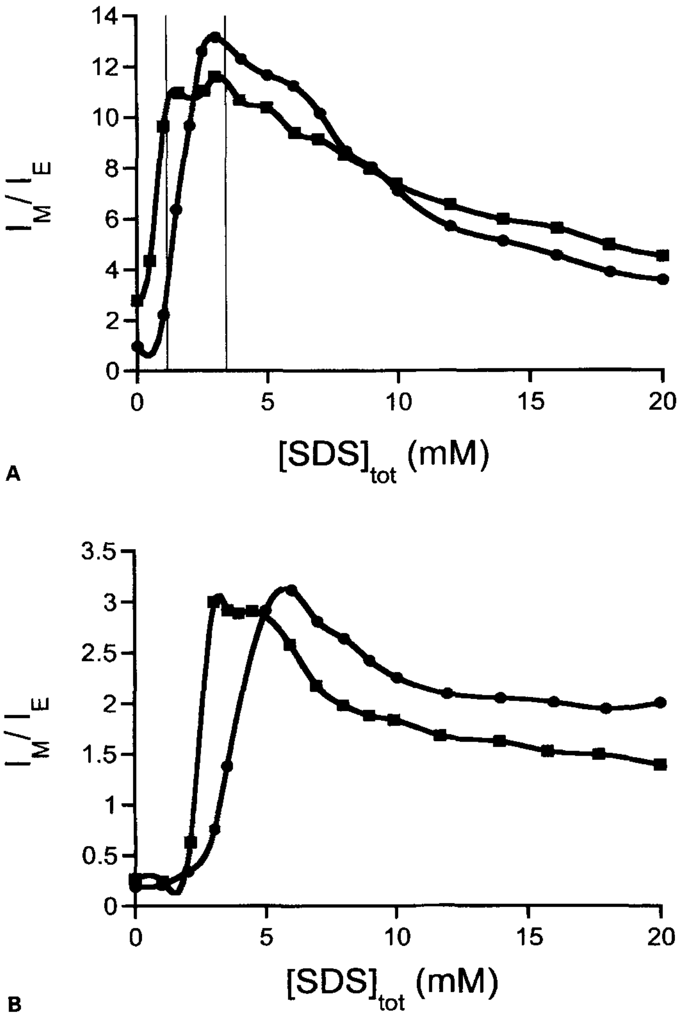


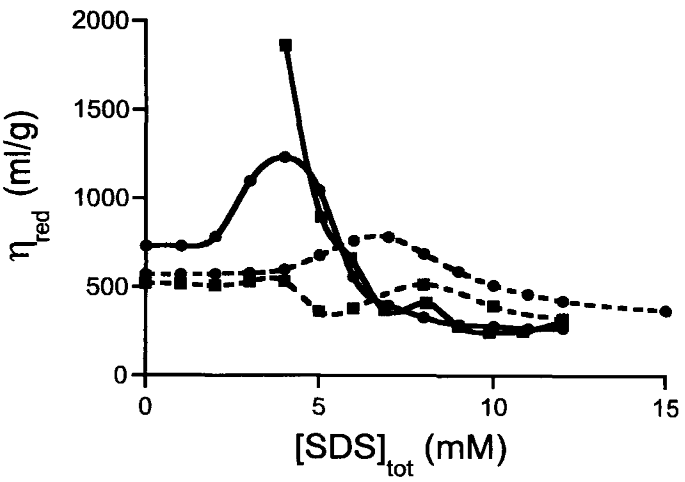
Fig. 4 The monomer to excimer intensity-ratio of 1,3-di(1-pyrenyl) propane, P3P, as a function of the total SDS concentration, $[SDS]_{tot}$, at 20 °C in 0.20% EHEC/water solutions. (●) no salt added; (■) 5 mM NaCl (a) EHEC-fraction-CST-103 (b) EHEC-fraction E230 G. Area between vertical lines in (a) indicates phase separation in presence of 5 mM NaCl

solution, I_M/I_E gives a maximum right after the composition corresponding to CAC, as seen in Fig. 4b. After the maximum I_M/I_E show a "shoulder" at $[SDS]_{tot} = 5$ mM similar to that of the salt-free CST-case, decreases and is lower than in the corresponding salt-free solution.

Viscosity

When SDS is added to a solution containing EHEC, the viscosity is altered as seen in Fig. 5. Similar observations have been made by others [17, 51]. This change in rheological behavior coincides with the onset of binding of SDS to both EHEC fractions. The reduced viscosity for CST solutions in the absence of salt is not altered at low SDS concentrations, but in a region beginning at approximately 2 mM SDS, η_{red} increases markedly with added SDS. At higher SDS concentrations, at about 4.5 mM, the increase reaches a maximum followed by a substantial decrease in reduced viscosity before it settles for a low value at approximately 8 mM. When salt is added to a CST solution with a sufficiently high SDS content, as to avoid phase separation [16], the maximum in η_{red} is increased although at SDS concentrations above 6 mM, the reduced viscosity is hardly affected. The EHEC fraction with a higher CP, E230 G, shows a similar behavior in salt-free conditions, although the rise in η_{red} is not as marked as it is for CST, and occurs at higher SDS concentrations. In the presence of salt, however, E230G behaves quite differently compared to CST. Upon increasing the concentration of salt, the reduced viscosity for the polymer solution gradually decreases for all SDS concentrations. When the SDS concentration is 5–6 mM the curve shows an inflection instead of a maximum. It should be noted that in this composition the polymer ih on the verge of phase separation [16].

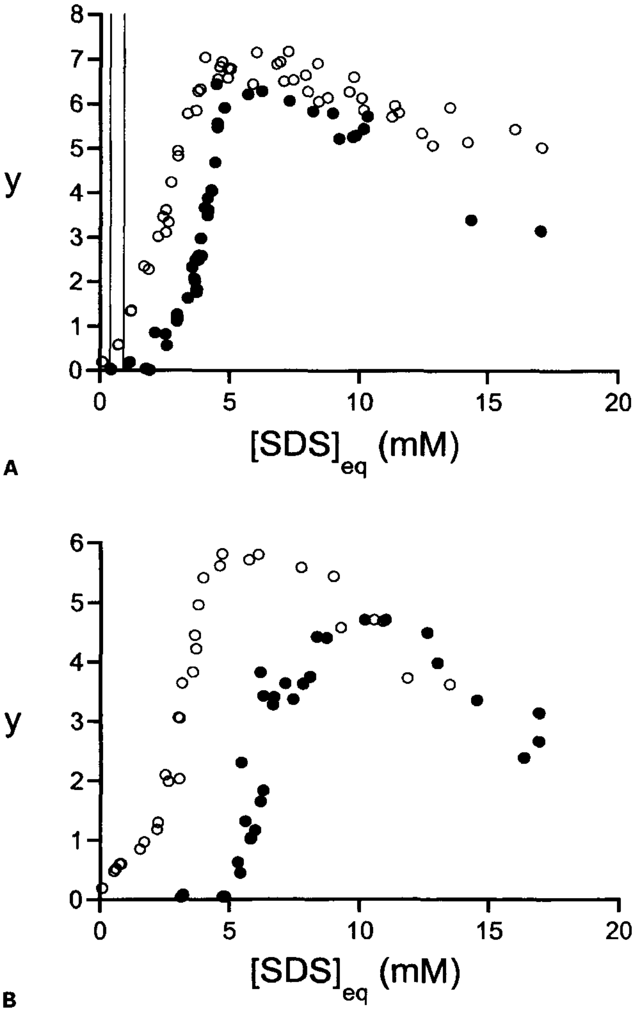
Fig. 5 The reduced specific viscosity, η_{red}, of 0.20% EHEC/water solutions as a function of the total SDS concentration, $[SDS]_{tot}$, at 20 °C (●) no salt added; (■) 5 mM NaCl. Full line, EHEC-fraction CST-103; dotted line, EHEC-fraction E230 G



Equilibrium dialysis

Concerning the SDS/CST system without salt, the adsorption isotherm displays the following features, as seen in Fig. 6a. At low SDS concentrations there is no adsorption at all, and the onset of binding occurs at $[SDS]_{eq} = 2.5$ mM, CAC, followed by a increase in y indicating

Fig. 6 Adsorption isotherms of SDS adsorbed to 0.20% EHEC. Millimoles of SDS bound per gram of EHEC, y, as a function of the equilibrium SDS concentration, $[SDS]_{eq}$, at 20 °C. (●) no salt added; (○) 5 mM NaCl. (a) EHEC-fraction CST-103, (b) EHEC-fraction E230 G. Area between vertical lines in (a) indicates phase separation in presence of 5 mM NaCl



a cooperative mechanism [8]. Between the CAC and the maximum in y a break point, as discussed in previous papers [12, 13, 15], is present. When the maximum in y is reached, ordinary micelles are assumed to occur in the bulk solution as a result of saturation of adsorbed SDS to EHEC [13]. After the maximum there is a decline in the adsorption [14]. When salt is added to the CST/SDS solutions the main difference compared to salt-free conditions is that the foot point, CAC, as well as the maximum in y (which is slightly higher in the presence of salt) is shifted to lower SDS concentrations. There is also a composition range at 20 °C where phase separation occurs. The cooperativity does not seem to be affected by the addition of salt. In Fig. 6b the adsorption isotherms obtained for E230G can be seen, and the graph representing the salt-free case displays the same shape as for CST, although the foot point and the maximum in y occurs at $[\mathrm{SDS}]_{\mathrm{eq}} = 5$ and 8 mM, respectively. Also the maximum amount of SDS adsorbed to E230G is lower than that of CST. When salt is added to the E230G/SDS/water system, the adsorption isotherm shows a considerably lowering of CAC, i.e. the adsorption starts at once, followed by a slow progress, which after a break point between CAC and maximum in y, the adsorption rises steeply up to the maximum. After the maximum, which is much higher in the presence of salt, there is a decrease in adsorption as in the salt-free case, although not as abrupt.

Discussion

The two cellulose derivatives studied are two polymers with the same back-bone structure but with different substitution as well as distribution of substituents. This leads to a difference in the hydrophilic and hydrophobic balance within the polymers, resulting in diversities in coil–coil interaction. This may affect the amount of hydrophobic "binding sites" exposed to additives in solution, illustrated by, for instance, different rheological behavior of the polymers. Although the EHEC-fractions show similarities in their interactions with the amphiphile SDS [16], the response to added-salt differs between the polymer/surfactant/water systems. It is found that NaCl affects the properties of E230G/SDS/water solutions to a higher degree than CST/SDS/water. It is also found that the macroscopic rheological properties of the salt-free EHEC/SDS/water system correlates well with the microscopic behavior on a molecular level, and the model of interaction proposed earlier [9–13, 15, 16] is supported.

When discussing the influence of salt on polymer/surfactant systems, one may begin with the effects of salt on solutions of pure surfactant and pure polymer, respectively. CST, like most uncharged polymers, is not affected by small amounts of salt [25], but E230G show an altered hydrodynamic volume as well as CP upon addition of 5 mM of salt [16]. The sensitivity of E230G to added salt has been reported by Nilsson et al. [25]. As for the surfactant SDS, the CMC is lowered by addition of salt, as seen in Fig. 1, where the hydrophobic index display a sharp decrease at CMC. The initial values in I_1/I_3 is somewhat raised when 5 mM NaCl is added to the system. This is probably due to the increased polarity of the solvent as sensed by pyrene below CMC. This effect can also be detected for the EHEC/SDS/NaCl/water systems in Figs. 2a and b. The lowering of CMC has also been verified by others [20, 50].

Let us first consider the more hydrophobic EHEC fraction CST 103. The onset of interaction between surfactant and polymer, at the CAC in terms of cluster formation, can be detected as a decrease in I_1/I_3 and is shown in Fig. 2a. For the salt-free CST/SDS/water system CAC, as sensed by the probe, is somewhat lower but still correlates fairly well to the CAC value given by the equilibrium dialysis data. This may be explained by the response of I_1/I_3 to the molecular changes, which occur at slightly lower amphiphile concentrations, than other techniques [27, 52]. This may be caused by the high sensitivity and high preferential distribution coefficient of the probe used. When salt is added to the CST/SDS/water solution there is a lowering in the sudden drop in hydrophobic index with respect to the SDS concentration, showing a CAC that corresponds to the onset of redistribution, as seen in Fig. 6a. I_1/I_3 also shows that the probe senses a more hydrophobic environment at SDS concentrations at or above CAC in presence of salt, which might be an indication of a decrease in the electrostatic repulsion between surfactant monomers and hence, more closely packed micelles (with respect to SDS) with a better shielding of pyrene from water. The hydrophobic index shows a slight decrease even after CAC, indicating a less polar environment for the probe as the clusters grow in size.

The average aggregation numbers, N_{p}, for CST, as seen in Fig. 3a, show a CAC similar to that of the adsorption isotherm. The decrease in N_{p} after the maximum is most probably an effect due to the small decrease in y at this region of SDS concentration. When salt is added to a CST solution with an SDS concentration high enough to prevent phase separation, i.e. 4 mM, N_{p} is higher than for salt-free conditions up to the point $[\mathrm{SDS}]_{\mathrm{tot}} = 12$ mM, where suddenly N_{p} for CST/SDS/water solutions in absence of salt has a larger average aggregation number. Since a corresponding change in adsorption is not found (more SDS is adsorbed at all concentrations in the presence of salt), this leads to the conclusion that the number of micelles increase on the polymer instead. The reason for this is unclear and needs to be looked into in more detail, but it may indicate a shift in coil–coil interaction that results in a difference in availability of hydrophobic binding sites.

The average aggregation numbers of polymer-bound clusters can also be related to the microviscosity index I_M/I_E. In a recent publication on the microviscosity of the salt-free CST/SDS/water system [15], a correlation was seen between the relative polymer content of the polymer–surfactant clusters and I_M/I_E. The maximum in I_M/I_E corresponds to very low-average aggregation numbers ($N_p \approx 10$), and similar effects are reported by others [53–56] for related polymer/surfactant systems. As more SDS is added, the clusters grow in size with respect to N_p, the relative polymer content decreases within the clusters, and the microviscosity index decreases. The maximum in I_M/I_E right after the CAC for the CST/SDS/water system, as can be seen in Fig. 4a, probably reflects clusters with a relatively high polymer content, giving rise to a high microrigidity due to the many covalently attached polymer segments constituting such a cluster [15]. These segments seem to hinder the excimer-formation of P3P to a higher degree than ordinary SDS micelles do, which is in analogy with similar findings by others [26, 40, 54, 57, 58]. When salt is added to the CST/SDS/water solutions, I_1/I_3 and N_p show that the interaction between polymer and surfactant starts at very low surfactant concentrations as can be seen in Figs. 3a and 2a, which correlates with the lowering of the abrupt increase in I_M/I_E on the SDS scale. There is an increase in I_M/I_E towards a plausible maximum, which is impossible to discuss since phase separation occurs. After the point of phase separation, I_M/I_E decreases gradually as the SDS concentration increases, which is compatible with the growing clusters, where the probe senses a less rigid environment. At about $[SDS]_{tot} = 10$ mM, there is a cross over between the lines where the microviscosity index becomes higher in the presence of salt than in the absence, the corresponding point of change can be found in N_p, which at this SDS concentration becomes lower in the system containing salt, thus, providing a higher viscosity for the P3P probe. It is interesting to note, that in the presence of salt at $[SDS]_{tot} = 12$ mM, N_p decreases and the microviscosity index increases, as the adsorption increases, all compared to the salt-free case. This leads to the conclusion that the surfactant is somehow redistributed so that the clusters become more and smaller along the chain.

The E230G/SDS/water system shows characteristics that differs as well as are similar to that of CST. With its higher CP, E230G can be expected to show a hydrophobic index indicating a CAC higher than that of CST, as seen in Fig. 2b. The I_1/I_3 curves for the E230G/SDS/water system with and without salt display more or less the same features as for CST. When it comes to average aggregation numbers, the E230/SDS/water system shows a resemblance to that of CST/SDS/water system, both systems with no salt present. The cluster formation starts at the same CAC as shown for equilibrium dialysis, N_p increases although the aggregates never grow as big as the ones bound to CST. The E230G sample, on the other hand, display larger average aggregation numbers of polymer-bound clusters in presence of NaCl at all SDS concentrations compared to the salt-free case. As for CST, there is a major increase in I_M/I_E at the CAC in the case of E230G due to cluster formation giving a more rigid environment for P3P, after which the microrigidity decreases according to the growing size of the bound clusters. When 5 mM of NaCl is added to the E230/SDS/water system, I_M/I_E decreases above CAC with respect to the salt-free system and the effect is more pronounced for E230 G, which can be related to the fact that this fraction has the larger salt-induced increase in y and N_p of the two. P3P also senses the decrease in CAC with the SDS-concentration similar to that of pyrene. Considering the salt-free systems, the cluster-growth of CST 103 with respect to the SDS concentration is slower than that of E230 G, which is seen from the slopes of the curves. The latter forms larger clusters faster after the CAC. This, in combination with the higher hydrophobicity of CST which promotes the ability to form densely packed mixed micelles with respect to the polymer, might explain why CST gives a higher microviscosity index right after the CAC. CST actually gives three to four times more rigid clusters initially than E230 G. These differences in microviscosity index between CST and E230G as well as several other nonionic cellulose ether will be discussed more thoroughly in a forthcoming paper from this laboratory. Here, it is worth noting the sensitive correlation between I_M/I_E and N_p as already discussed, and between I_M/I_E and y as well: sudden changes in the derivatives of I_M/I_E and y coincide with respect to the SDS concentration as can be seen in Figs. 4a, b and 6a, b. For CST in the absence of salt, a break point occurs in the adsorption isotherm at $[SDS]_{eq} = 3.5$ mM coinciding with the shoulder in I_M/I_E at $[SDS]_{tot} = 7$ mM. The same situation appears for E230G in the presence of salt at $[SDS]_{eq} = 2.5$ mM and $[SDS]_{tot} = 5$ mM, respectively. These break points are believed to illustrate compositions at which SDS passes to adsorb to single strands of EHEC compared to partly self-aggregated EHEC at lower SDS concentrations [12].

Both EHEC fractions interact with SDS, under salt-free conditions, in a similar way but to different magnitudes, and both systems display different tendencies under the influence of salt. This reflects the differences in substitution of the polymers. The main features may be summarized from the present study in relation to previous results [16]. When SDS is added to a solution of CST, interaction occurs leading to continuous solubilization of the polymer, which is indicated by an increase in CP with added SDS. The more hydrophilic sample E230G experiences a lowering in CP at low SDS concentrations, a "salting-out" effect, due to the free ions before the interaction starts. To enhance the solubility of E230G a sufficient amount of SDS is required. The interaction means that SDS is redistributed to EHEC in the form of clusters on the polymer back-bone. This leads to an alteration of the rheological pattern of the polymer solution, which can be noticed as a change in reduced viscosity. The mechanism behind this behavior is probably alike for CST and E230G, which is supported by the following observations. When $y = 1$ (i.e. 1 mmol of SDS is adsorbed per gram EHEC) the compositions for both fractions correspond to maximum in bulk viscosity, maximum in hydrodynamic interaction as expressed by Huggins constant (k_H) and minimum in intrinsic viscosity [16]. At this point the polymer is not saturated with SDS and the cluster size is about $N_p = 17$ for both EHEC fractions. This is further supported by the values of microviscosity index indicating high microrigidity at these particular compositions. This leads to the conclusion that two or more polymer coils share one cluster which result in a three-dimensional network. This mechanism has been found to depend on polymer concentration [9, 11–13, 15, 16]. In the work on microviscosity in the salt-free CST/SDS/water system it was stated [15] that the coincidence between the maxima in I_M/I_E and viscosity with respect to the SDS concentration, a situation shown from these data to hold also for the E230G/SDS/water system, probably derives from two different mechanisms: the maximum in viscosity has its origin in a three-dimensional network as already mentioned, while the maximum in I_M/I_E corresponds to a composition with a low N_p indicating polymer dense clusters in accord with the discussion above. Raising the polymer concentration from below to above c^* result only in a minor increase in the maximum in I_M/I_E [15]. That is, I_M/I_E is not found to be sensitive to whether the mixed surfactant-polymer micelles are shared within the same polymer string or between many, as the viscosity obviously is. At higher SDS concentrations beyond the maximum in viscosity the driving force to share clusters at hydrophobic binding sites seems to disappear leading to a break down of the network, and an increased solubilization of polymer in normal micelles. This is in agreement with a recent paper by Piculell et al. [59] in which the EHEC/SDS/water system was studied in the semi-dilute regime by means of rheology and NMR self-diffusion and it was argued that the three-dimensional network breaks down as soon as SDS dominates the clusters.

When salt is added, both E230G and CST show a minimum in CP at the SDS concentration corresponding to the maximum in η_{red} for each particular polymer. This is in accord with the general model described above. As more SDS is added to the polymer solutions, "deaggregation" of the network occurs, resulting in an increased adsorption of SDS to the exposed binding sites, leading to a more solubilized polymer, which is less sensitive to added salt. On the whole, at high concentration of SDS, the effect of 5 mM NaCl diminish and the properties of EHEC/SDS/NaCl water system approach that of the salt-free system. This is evident from the dialysis, bulk viscosity and average aggregation numbers of polymer-bound clusters. It may be a consequence of an increasing amount of hydrophobic parts of the polymer solubilizing in mixed polymer/surfactant micelles, thus, giving almost identical reduced viscosity as the polymer becomes more shielded from added salt, and at the higher SDS concentrations the contribution to the total ion-strength originating from 5 mM NaCl decreases.

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References

1. Bahadur P, Dubin P, Rao YK (1995) Langmuir 11:1951
2. Carlsson A (1989) Nonionic cellulose ethers-interactions with surfactants, solubility and other aspects, Dissertation. University of Lund
3. Saito S, Kitamura K (1971) J Colloid Interface Sci 35:346
4. Illiopolous I, Wang T, Audebert R (1991) Langmuir 7:617
5. Francois J, Dayantis J, Sabbadin J (1985) Eur Polym J 21:165
6. Breuer MM, Robb ID (1972) Chem Indust 13:530
7. Dubin P (ed) (1985) Microdomains in Polymer Solutions. Plenum Press, New York
8. Goddard ED, Ananthapadmanabhan KP (eds) (1993) Interactions of Surfactants with Polymers and Proteins. CRC Press, Boca Raton
9. Holmberg C, Nilsson S, Singh SK, Sundelöf LO (1992) J Phys Chem 96:871
10. Nilsson S, Holmberg C, Sundelöf LO (1994) Colloid Polym Sci 272:338
11. Nilsson S, Holmberg C, Sundelöf LO (1995) Colloid Polym Sci 273:83
12. Evertsson H, Nilsson S, Holmberg C, Sundelöf LO (1996) Langmuir 12:5781
13. Holmberg C, Sundelöf LO (1996) Langmuir 12:883
14. Holmberg C, Nilsson S, Sundelöf LO (1997) Langmuir 13:1392
15. Evertsson H, Nilsson S (1997) Macromolecules 30:2377
16. Holmberg C (1996) Colloid Polym Sci 274:836

17. Carlsson A, Karlström G, Lindman B (1986) Langmuir 2:536
18. Karlström G, Carlsson A, Lindman B (1990) J Phys Chem 94:5005
19. Horin S, Arai H (1970) J Colloid Interface Sci 32:547
20. Murata M, Arai H (1973) J Colloid Interface Sci 44:475
21. Singh K, Deepak S, Bahadur P (1979) Colloid Polym Sci 257:57
22. Kalyanasundaram K (1987) Photochemistry in Microheterogeneous Systems. Academic Press, Orlando
23. Grieser F, Drummond CJ (1988) J Phys Chem 92:5580
24. Winnik FM, Regismond STA (1996) Colloids Surf A 118:1
25. Nilsson S, Sundelöf LO, Porsch B (1995) Carbohydr Polym 28:265
26. Winnik FM, Winnik MA, Ringsdorf H, Venzmer J (1991) J Phys Chem 95:2583
27. Kalyanasundaram K, Thomas JK (1977) J Am Chem Soc 99:2039
28. Turro NJ, Baretz BH, Kuo PL (1984) Macromolecules 17:1321
29. Zana R, Lianos P, Lang J (1985) J Phys Chem 89:41
30. Nilsson S (1995) Macromolecules 28:7837
31. Turro NJ, Yekta A (1978) J Am Chem Soc 100:5951
32. Witte FM, Engberts JBFN (1989) Colloids Surf 36:417
33. Moroi Y, Humphry-Baker R, Grätzel M (1987) J Colloid Interface Sci 119:588
34. Croonen Y, Gelade E, Zegel MVd, Auweraer MVd, Vandendrlessche H, Schryver FCD, Almgren M (1983) J Phys Chem 87:1426
35. Zachariasse KA (1978) Chem Phys Lett 57:429
36. Georgescauld D, Desmasèz JP, Lapouyade R, Babeau A, Richard H, Winnik M (1980) Photochem Photobiol 31:539
37. Lianos P, Lang J, Strazielle C, Zana R (1982) J Phys Chem 86:1019
38. Parthasarathy R, Labes MM (1990) Langmuir 6:542
39. Miyagishi S, Suzuki H, Asakawa T (1996) Langmuir 12:2900
40. Yekta A, Duhamel J, Brochard P, Adiwidjaja H, Winnik MA (1993) Macromolecules 26:1829
41. Nivaggioli T, Tsao B, Alexandridis P, Hatton TA (1995) Langmuir 11:119
42. Zana R (1987) Surfactant Solutions: New Methods of Investigation. Marcel Dekker, New York
43. Zachariasse KA, Vaz WLC, Sotomayor C, Kühnle W (1982) Biochem Biophys Acta 688:323
44. Hare F, Lussan C (1977) Biochim Biophys Acta 467:262
45. Almgren M, Löfroth J-E (1981) J Colloid Interface Sci 81:486
46. Malliaris A (1987) Progr Colloid Polym Sci 73:161
47. Barr G (1931) A Monograph of Viscometry. Oxford University Press, London
48. Fischman ML, Eirich FR (1971) J Phys Chem 75:3135
49. Shirahama K (1974) Colloid Polym Sci 252:978
50. Hall DG, Tiddy GJT (1981) Anionic Surfactants. Marcel Dekker, New York
51. Wang G, Olofsson G (1995) J Phys Chem 99:5588
52. Ananthapadmanabhan KP, Goddard ED, Turro NJ, Kuo PL (1985) Langmuir 1:352
53. Anthony O, Zana R (1996) Langmuir 12:1967
54. Anthony O, Zana R (1996) Langmuir 12:3590
55. McGlade MJ, Randall FJ, Tcheurekdjian N (1987) Macromolecules 20:1782
56. Winnik FM, Ringsdorf H, Venzmer J (1991) Langmuir 7:905
57. Aizawa M, Komatsu T, Nakagawa T (1977) Bull Chem Soc Jpn 50:3107
58. Chu DY, Thomas JK (1991) Macromolecules 24:2212
59. Piculell L, Thuresson K, Ericsson O (1995) Faraday Discuss 101:307