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A thermosensitive carrageenan-based polymer: Synthesis, characterization and interactions with a cationic surfactant

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ABSTRACT

Novel polyelectrolytes were obtained by grafting N-isopropylacrylamide (NIPAM) on the ι-carrageenan (CAR) chain. Two polymers with different grafting degrees were synthesized. The polymers were found to show the lower critical solution temperature (LCST) close to that of PNIPAM. The LCST values were dependent on the concentration of salt and cationic surfactant. The interactions of CAR-*graft*-PNIPAM with a model cationic surfactant-dodecyltrimethyl ammonium chloride (DTAC) in water and 0.15 M NaCl were studied. It was found that both ι-carrageenan and CAR-*graft*-PNIPAM polymers interact with DTAC. The presence of CAR-graft-PNIPAM in the solution of DTAC induces formation of surfactant aggregates at the critical aggregation concentration much lower than the cmc of the surfactant. Cac increased with ionic strength. The values of cac for CAR-*graft*-PNIPAM – DTAC system and standard free enthalpy changes attributed to the complexation process were determined. The results obtained for CAR-*graft*-PNIPAM were compared with these for the non-modified ι-carrageenan. The surfactant interactions with non-modified and grafted polymers were found to be different in nature.

Introduction

Recently, there is a growing interest in the studies on interactions between polymers (Dias, Mel'nikov, Lindman, & Miguel, 2000; Hansson, 2006; Kogej, 2010; Pan et al., 2012; Santos et al., 2010), polysaccharides in particular (Desbrieres, Bousquet, & Babak, 2010; Kogej, 2008; Singh et al., 2008), and surfactants. This interest is driven by the fact that these subtle in their nature interactions have profound effect on the solution properties of the individual components. The systems in which stimuli-responsive polymers (smart polymers) are used belong to these, attracting special attention (Ahn, Ahn, & Song, 2008; Chen, Katsuyama, Gong, & Osada, 2003; Chiper, Fournier, Hoogenboom, & Schubert, 2008; Ganji & Abdekhodaie, 2008; Kim, Gong, & Osada, 1999; Kwon, Osada, & Gong, 2006; Shen et al., 2008; Tokuyama & Kato, 2008). They are widely studied because of their potential practical applications as drug delivery systems, and polymer supports for catalysts, chromatography column packing or cell and tissue culture. Poly(N-isopropylacrylamide) (PNIPAM), a thermosensitive polymer, which in aqueous solution displays lower critical solution temperature (LCST), is among the most frequently studied polymers. PNIPAM homopolymer exhibits thermoreversible phase separation in the aqueous solution in the temperature range of 31–35 °C (Heskins & Guillet, 1968). It was observed that the copolymers of NIPAM with various monomers are also thermosensitive and the value of the LCST is strongly dependent on their composition. This observation is of great importance as it suggests the way for the fabrication of the polymers with a predetermined range of thermosensitivity, which can serve defined purposes. The other method which can be used to tune the thermal sensitivity of these polymers is their interactions with a surfactant (Kogej, 2008; Singh et al., 2008).

Most of the studies on the NIPAM-based polymers are carried out with synthetic systems. However, for many applications, e.g. biomedical, the systems of natural origin would be preferable. In that view, we have previously studied the derivatives of hydroxypropylcellulose, a natural, thermosensitive polymer (Rosół, Szczubiałka, Jachimska, Zapotoczny, & Nowakowska, 2008; Szczubiałka, Rosół, & Nowakowska, 2006).

This paper presents the results of our work on the novel thermosensitive polymer which is based on carrageenan. Carrageenans are a family of linear sulphated polysaccharides–galactose derivatives. They are composed of alternating 3-linked β-D-galactopyranose and 4-linked α-D-galactopyranose or 4-linked 3,6-anhydro-α-D-galactopyranose, forming the disaccharide repeating unit of carrageenans. There are three main classes of carrageenans: kappa, iota and lambda. The sulphated galactans are classified

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according to the presence of the 3,6-anhydro-bridge and the number of sulphate groups. (Campo, Kawano, Brazda Silva, & Carvalho, 2009). For kappa carrageenan charge/dimer ratio is about 1 and anhydrogalactose/dimer ratio is also 1. It has ability to form strong and rigid gels. Iota carrageenan forms soft gels and has charge/dimer ratio about 2.0 and anhydrogalactose/dimer ratio about one. Lambda carrageenan's charge/dimer ratio is about 3.0 and it does not have anhydrogalactose units in its structure. Carrageenans having anhydrogalactose units are able to form helical structures and aggregate forming gels. Lambda carrageenan which does not have anhydrogalactose units does not form gels (Campo et al., 2009). Because of the ability to undergo gelation these polysaccharides are widely used in the food and other industries as thickening and stabilizing agents (Kwon et al., 2006).

We have chosen iota carrageenan for our studies because it is characterized by the relatively high value of charge/dimer ratio and its sulphate groups are oriented toward the external part of the macromolecule, thus making easier the interactions with charged species of other types present in the system. We have modified iota carrageenan (ι-carrageenan) by grafting NIPAM chains on its polysaccharide backbone. This was done to obtain a natural polymer modified as little as possible, however, with the degree of modification with PNIPAM high enough to show thermosensitivity (i.e. LCST). This is of practical importance since such a polymer or a complex of such a polymer with a surfactant may be easily precipitated from the solution just by heating it above LCST allowing removal of a surfactant from water. Thus, such a polymer may be potentially applied in the purification of water from surfactants.

Experimental

Materials

ι-Carrageenan (CAR, Fluka), N-isopropylacrylamide (NIPAM, Aldrich, 99%), dodecyltrimethylammonium chloride (DTAC, Fluka, 99%), potassium permanganate (POCH Gliwice, analytical grade), sodium hydroxide (POCH Gliwice, analytical grade), sulfuric acid (POCH Gliwice, analytical grade) were used as received. Water was distilled twice. Pyrene (Aldrich, 98%) was recrystallized twice from methanol.

Polymer synthesis

The typical polymer synthesis procedure was as follows. In a 250-ml three-necked flask 3 g of ι-carrageenan was dissolved in 200 ml of water. The solution was degassed by bubbling with nitrogen for 30 min and 0.0077 g (0.5 mM) of $KMnO_4$ dissolved in 2 ml of water was added. After 5 min the solution became colorless and 1.137 g (12 mM) of H_2SO_4 and 1.599 g (14 mM) of N-isopropylacrylamide (NIPAM) was added. Then the reaction mixture was heated and kept at 60 °C for 4 h. The mixture was stirred constantly with a magnetic stirrer and bubbled with nitrogen. The reaction was terminated by rapid cooling of the reaction mixture and neutralizing with NaOH solution. The precipitate was removed by decantation. The polymeric solution was dialyzed for 1 week against distilled water and freeze-dried.

Apparatus

The elemental analysis (C, H, and N) was performed with an Euroea 3000 elemental analyzer. GPC analyses were performed using a Waters chromatographic system equipped with a Waters Ultrahydrogel Linear column and a Waters 2996 UV–Vis Photodiode Array (PDA) Detector. The 0.1 M NaCl aqueous solution was used as an eluent and the flow rate was 1 ml min^{-1}. ^1H NMR spectra recorded in D_2O, at ambient temperature on a Bruker 500 MHz spectrometer. FT-IR spectra were recorded using a Bruker Equinox 55 spectrophotometer. Fluorescence spectra of pyrene were measured using an SLM-AMINCO spectrofluorimeter. The excitation wavelength was adjusted at λ_{ex} = 320 nm. The spectra were corrected for the apparatus response using a function supplied by the manufacturer. Surface tension was measured using a K9 Krüss tensiometer. Isothermal Titration Calorimetry was performed using a VP-ITC isothermal titration calorimeter, MicroCal Inc., Northampton, MA, USA. The amount of heat absorbed or emitted was calculated using an Origin software supplied by the manufacturer. The baseline was generated automatically and at low signal/noise ratio, manually corrected. For all the experiments reference measurements were performed and surfactant solution was added to water. Referential thermal effect was subtracted from thermal effects obtained in the presence of the polymer.

LCST measurements

The LCST values for the polymers were measured using a Hewlett-Packard 8452A spectrophotometer equipped with a Hewlett-Packard 89090A Peltier temperature control accessory, as described earlier (Nowakowska, Szczubiałka, & Grębosz, 2004). In short, the solution was heated with the Peltier accessory within the range 15–70 °C. The solution was heated at the rate of about 0.5 °C min^{-1} and stirred at the rate of 5 s^{-1}. The LCST values were determined from the changes in the turbidity with temperature, expressed as $1 - T$, where T was apparent transmittance of the polymer solutions at 400 nm.

DLS measurements

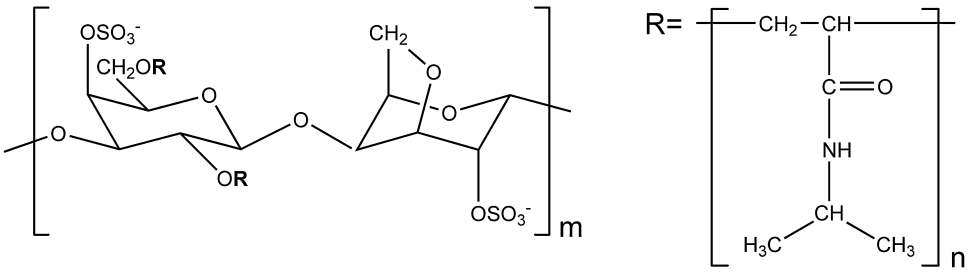
Dynamic light scattering was measured using a Malvern Nano ZS (Malvern Instruments Ltd., Worcestershire, UK) and polymer solutions at 2 g/L. The Nano ZS instrument incorporates non-invasive backscatter (NIBSTM) optics. DLS technique measures the time-dependent fluctuations in the intensity of scattered light which occur because the particles are undergoing Brownian motion. Analysis of these intensity fluctuations enables the determination of the diffusion coefficients of the particles which are converted into a size distribution. The time-dependence autocorrelation function of the photocurrent was acquired every 10 s, with 15 acquisitions for each run. The sample of solutions was illuminated by a 633 nm laser, and the intensity of light scattered at an angle of 173° was measured by an avalanche photodiode. The z-average diameter (dz) and the polydispersity index (PD) of the samples were automatically provided by the instrument using cumulant analysis. Rh data are apparent values obtained via the Stokes–Einstein equation for spheres.

Solubilization

Pyrene used in the fluorescence spectroscopy studies was solubilized in the systems by slowly injecting microliter volumes of a probe in methanol into the aqueous solutions of the polymers under vigorous shaking. The solutions were shaken for at least 10 min and left in the dark to equilibrate for at least 2 h.

Results and discussion

The grafting of PNIPAM on ι-carrageenan was carried out using $KMnO_4$ as a redox initiator. Such a procedure was previously used for grafting acrylonitrile onto starch (Hebeish, El-Thalouth, El-Kashouti, & Fattah, 1979). It was suggested that the initiation process occurs through the reduction of Mn^{+4} to Mn^{+3} and/or to Mn^{+2} and that the radicals are formed only along the polysaccharide chain so the formation of the PNIPAM homopolymer is excluded



Scheme 1. Structure of CAR-*graft*-PNIPAM.

(Kim et al., 1999). The general structure of the CAR-*graft* -PNIPAM polymer is proposed in Scheme 1.

Polymers with two different degrees of substitution were obtained. The degree of substitution of CAR-*graft*-PNIPAM polymers, defined as the average number of NIPAM mers per the number of monosaccharide units, i.e. the number of NIPAM units attached to a galactopyranose unit of carrageenan, was estimated based on the results of elemental analysis. It was found to be 0.065 and 0.75 which means that statistically there is 65 and 750 mers of NIPAM per 1000 galactopyranose units for CAR-*graft*-PNIPAMI and CAR-*graft*-PNIPAMII respectively.

The CAR-*graft*-PNIPAM polymers were characterized using NMR, FT-IR, GPC and elemental analysis. ^{1}H NMR spectra of the graft polymer (data not shown) display signals at 1.14 ppm characteristic of the methyl group in PNIPAM, while the signals at 1.57 and 2.01 ppm are characteristic of chain protons in PNIPAM grafts. These signals are absent in the NMR spectrum of the non-modified ι-carrageenan confirming successful grafting reaction. Fig. 1 shows the GPC traces of both non-modified and grafted ι-carrageenan. Since the initiator redox system applied was based on potassium permanganate, it may be expected that the grafting reaction may result in the oxidation of hydroxyl polysaccharide groups to carbonyl groups (Hebeish et al., 1979). In fact, FT-IR spectrum of the grafted carrageenan (data not shown) shows a very weak band at 1720 cm^{-1}, which can be due to the carbonyl groups formation. However, taking into account that the carbonyl stretching vibration gives very intense bands it may be concluded that the oxidation reaction, if any, is negligible and does not influence the properties of the polymer synthesized. GPC chromatogram obtained for non-modified carrageenan has shown a major peak at the retention time of about 16 min. There are also two small peaks at a shorter and a longer retention time, respectively. This indicates that the non-modified ι-carrageenan contains one major fraction and two small fractions of higher and lower molecular weight, respectively. This can be expected for the natural polymer which usually is a mixture of macromolecules of various sizes and molecular masses. The chromatogram for the grafted polymer, CAR-*graft*-PNIPAMI, shows basically only one peak, at the retention time of about 25 min. Higher retention time for CAR-*graft*-PNIPAM than for ι-carrageenan may suggest that the macromolecules adopt a conformation corresponding to a smaller hydrodynamic radius than that of the non-modified ι-carrageenan. This can be explained considering that the grafting of PNIPAM results in an increase of hydrophobicity of the polymer chains and changes in their interactions with water. The shift is also expected to originate from the partial degradation of ι-carrageenan chains that may be concomitant to the grafting of polyNIPAM. It has been shown that ι-carrageenan can partially degrade at the acidic conditions comparable to those present in the reaction vessel (Hjerde, Smidsrod, & Christensen, 1996; Hjerde, Smidsrod, Stokke, & Christensen, 1998; Karlsson & Singh, 1999).

Thermosensitivity of the obtained polymers was studied based on the turbidity measurements. It was found that CAR-*graft*-PNIPAM polymers are thermosensitive. The lower critical solution temperature (LCST) for CAR-*graft*-PNIPAMI and CAR-*graft*-PNIPAMII is 35 and 34 °C, respectively (in an aqueous solution, polymer concentration c_p = 5 g/L). This is in agreement with a general rule that copolymerization of NIPAM with a hydrophilic monomer or grafting on a hydrophilic polymer increases its LCST (Hirose & Shibayama, 1998; Nowakowska, Szczubiałka, & Grębosz, 2003; Rosół et al., 2008; Szczubiałka et al., 2006). This phase transition is sensitive to the ionic strength of the solution – an increase in the ionic strength of the system decreases the LCST value (Fig. 2). For example, in the presence of NaCl at the concentration of 0.6 M, the LCST of CAR-*graft*-NIPAMI was found to be 27 °C as found from the turbidity measurements. This can be explained considering the shielding of the electrostatic repulsion between the ionic sulphate

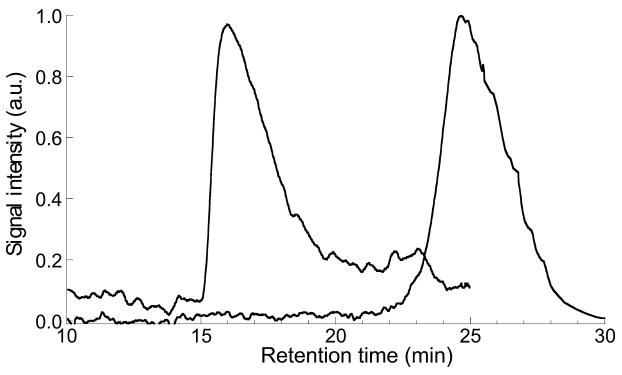


Fig. 1. GPC chromatograms for non-modified (left) and grafted (right) CAR (eluent: 0.1 M NaCl c_p = 1 g/L).

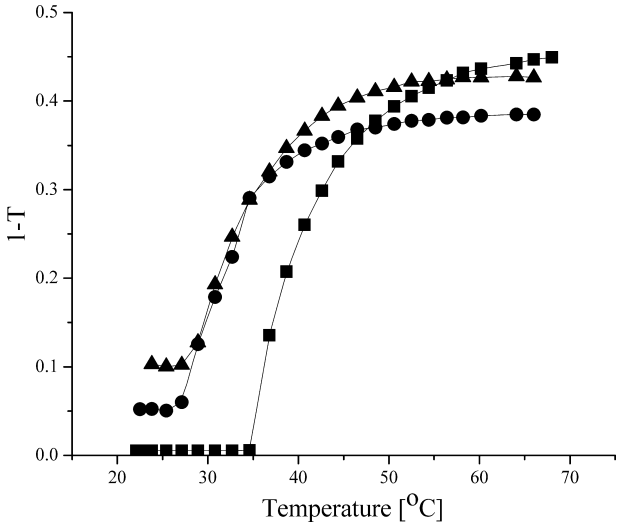


Fig. 2. Turbidity vs. temperature for CAR-*graft*-PNIPAMI aqueous solution in a presence of various salt (NaCl) concentrations: 0 (■); 0.6 (●); and 0.8 (▲) mol/dm^3, c_p = 5 g/L.

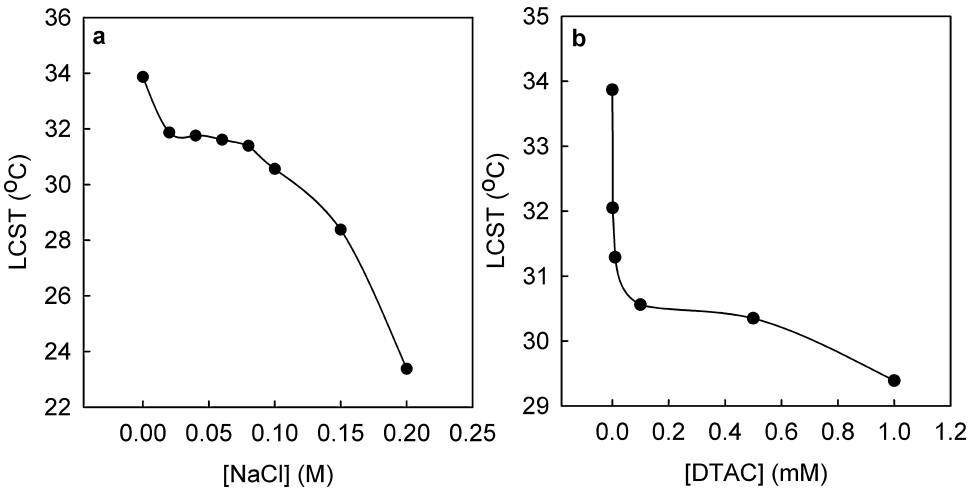


Fig. 3. The dependence of LCST of CAR-*graft*-PNIPAMII on (a) ionic strength and (b) [DTAC], c_p = 0.1 g/L.

groups present in the ι-carrageenan structure by sodium counterions which results in a decreased solubility of the system. For CAR-*graft*-PNIPAMII the influence of increased ionic strength on the LCST (Fig. 3a) was compared with the influence of the presence of different concentrations of a cationic surfactant DTAC (Fig. 3b). The LCST of CAR-*graft*-PNIPAMII was found to decrease from 34 °C in water to 29.3 °C in 1 M NaCl for the polymer concentration c_p = 0.1 g/L. The presence of DTAC in the polymer solution resulted in the analogical however more pronounced decrease of LCST. 1 mM [DTAC] induced the same changes in LCST of CAR-*graft*-PNIPAMII as around 0.15 M NaCl. This suggests that in the case of surfactant shielding is not the only one mechanism responsible for lowering LCST. Polymer–surfactant interactions decrease LCST even stronger (Nowakowska et al., 2003).

The change in the temperature from below the LCST to the values exceeding LCST is known to affect hydrogen bonding in water-PNIPAM system. This in turn should influence the size of polymeric coils. To find out how temperature affects the dimensions of the objects formed by the polymer in the solution dynamic light scattering measurements were performed for CAR-*graft*-PNIPAMI (Fig. 4), although taking account on the fact that the CAR chains are stiff and their shape is far from spherical, the R_h values obtained should be treated rather as relative than absolute ones. It was found that increasing the temperature from 25 °C (i.e. below LCST) to 40 °C (above the LCST) results in the increase of the hydrodynamic radius of the objects in the grafted carrageenan solution from 12 nm to about 110 nm which could be interpreted as resulting from the formation of the intermolecular aggregates which is typical for PNIPAM at temperatures higher than LCST. Small R_h of the objects below LCST also excludes networking due to chain termination via recombination. Further increase of turbidity at temperatures above 40 °C (Fig. 2) indicates that the size of aggregates still increases to values much greater than 110 nm. This allows their easy removal by simple filtration or centrifugation.

The interactions between ι-carrageenan and CAR-*graft*-PNIPAM with a cationic surfactant, dodecyltrimethyl ammonium chloride (DTAC), were investigated using three different, complementary physicochemical techniques: steady-state fluorescence measurements, surface tension measurements, and isothermal titration calorimetry.

In order to find the values of critical aggregation concentration (cac) for the surfactant in the presence of the polymers studied, we have applied pyrene, a fluorescent molecular probe. This probe is sensitive to the polarity of the environment in which it resides. Namely, the ratio of the intensity of the third and first vibrational bands, I_3/I_1, in the fluorescence emission spectrum can serve as a measure of the polarity of the medium surrounding the probe molecule, being low in the polar media and high in the hydrophobic environment (Kalyanasundaram & Thomas, 1977). Fig. 5 shows the plots of I_3/I_1 for pyrene solubilized in the ι-carrageenan – DTAC and CAR – *graft*-PNIPAMI-DTAC at a constant polymer concentration (c = 0.1 g/L) and various DTAC concentrations. Dependence of I_3/I_1 for DTAC solutions on the surfactant concentrations was also included.

One can observe that the I_3/I_1 ratio for the DTAC aqueous solutions is constant in the surfactant concentration range of 10^{-6} to 10^{-2} mol/L, and equal to 0.59 which is characteristic of water.

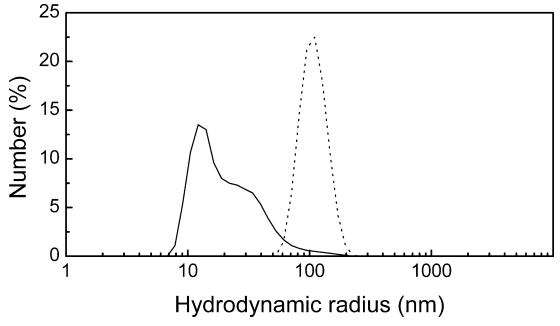


Fig. 4. Size of the objects in 2.0 g/L solution of CAR-*graft*-PNIPAMI at 25 °C (solid) and at 40 °C (dotted).

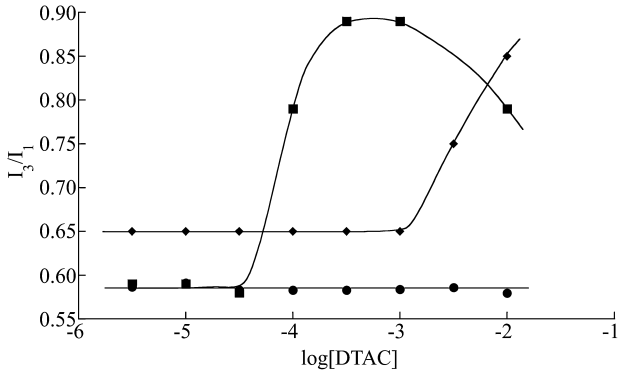


Fig. 5. The dependence of I_3/I_1 ratio of pyrene on DTAC concentration (mol/dm^3) in aqueous solution of DTAC (●), ι-carrageenan-DTAC (■), and CAR-*graft*-PNIPAMI – DTAC (♦) at c_p = 0.1 g/L.

Table 1
Critical aggregation concentrations, standard free enthalpy values for the systems of CAR–DTAC and CAR-*graft*-PNIPAM – DTAC at c_p = 0.1 g/L and T = 25 °C and excess Gibbs energy for polymer-bound aggregates stabilization (T = 25 °C).

Polymer	cac (mol/L)	ΔG_a^o (kJ/mol)	ΔG (J/mol)
CAR	$3.2{\cdot}10^{-5}$	−25.6	16.0
CAR-*graft*-PNIPAM	$3.2{\cdot}10^{-3}$	−14.2	4.6

This indicates that pyrene molecules are surrounded by water and ensures that the experiment is carried out under the conditions at which the surfactant micellization is excluded. These findings are in agreement with the literature data – according to the literature, the value of critical micellization concentration (cmc) for DTAC was determined to be 2.03×10^{-2} M (Jönsson, Lindmann, Holmberg, & Kronberg, 1998). For the ι-carrageenan – DTAC and CAR-*graft*-PNIPAMI – DTAC systems the values of I_3/I_1 increase rapidly at certain concentrations of DTAC, indicating the onset of the formation of hydrophobic domains. The cac of the surfactant can be determined by the extrapolation of two parts of the curves presented in Fig. 5. The values of the cac are given in Table 1. One can notice that the values differ considerably; the surfactant aggregation at the non-modified ι-carrageenan chains is effective at very low concentration, cac = 3.2×10^{-5} M, while for CAR-*graft*-PNIPAMI that process occurs at much higher concentration, cac = 3.2×10^{-3} M. Interestingly however, the solution of CAR-*graft*-PNIPAMI is somewhat more hydrophobic (I_3/I_1 = 0.65) than that of non-modified ι-carrageenan which shows I_3/I_1 very close to that characteristic of water. The PNIPAM side chains grafted on the ι-carrageenan backbone affect the polarity of the polymer, most likely by affecting hydrogen bonding between polymer and water.

The influence of ionic strength on CAR-*graft*-PNIPAMII-surfactant interactions was followed using the pyrene fluorescence. Changes of I_3/I_1 upon an increase of DTAC concentration in water and 0.15 M NaCl are shown in Fig. 6. I_3/I_1 at [DTAC] = 10^{-6} M is higher in water than in 0.15 M NaCl and both values correspond to I_3/I_1 in polymer solutions without surfactant added. Thus difference in hydrophobicity of the microenvironment should be ascribed to the polymer conformational changes upon increase of ionic strength. The critical aggregation concentration of DTAC on CAR-*graft*-PNIPAMII increased from around 10^{-5} M in water to $4.4{\cdot}10^{-4}$ M in 0.15 M NaCl.

The influence of temperature on CAR-*graft*-PNIPAM interactions with DTAC were also studied using the surface tension measurements (data not shown). It was found that increasing temperature resulted only in the vertical shift of the plots resulting from the decrease of the surface tension of water with temperature and no qualitative difference in the shape of the plots was observed.

Based on the cac values for the above systems and using the well known thermodynamic equation (Eq. (1)), the values of standard free enthalpy of aggregation, ΔG_a^o, (the free enthalpy change when

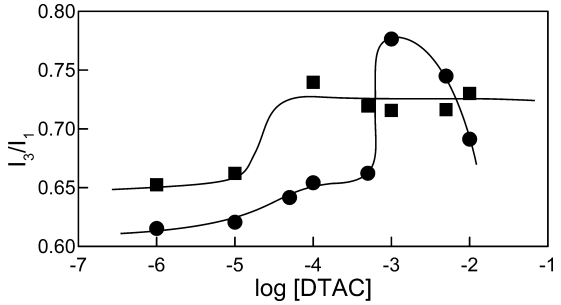


Fig. 6. The dependence of I_3/I_1 ratio of pyrene on DTAC concentration (mol/L) in water (■) and in 0.15 mol/L NaCl (●) at c_p = 1.0 g/L.

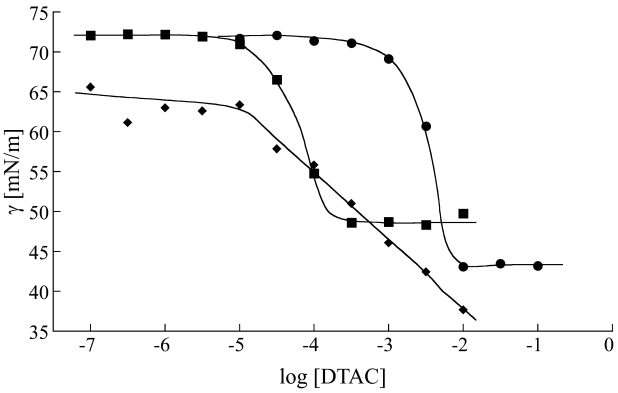


Fig. 7. The dependence of surface tension of aqueous polymer solutions (c_p = 0.1 g/L) on DTAC concentration: (●), CAR – DTAC (■), and CAR-*graft*-PNIPAMI – DTAC (♦).

the surfactant molecules are transferred from the aqueous phase to a polymer–DTAC aggregate) were estimated (Kogej & Škerjanc, 1999) (see Table 1):

$$\Delta G_a^o = RT \ \ln(\text{cac}) \tag{1}$$

where cac is the critical aggregation concentration; R, molar gas constant; T, absolute temperature.

The excess Gibbs energy that stabilizes the polymer–surfactant aggregates in comparison with the free micelles (at the temperature of 25 °C) was calculated using the equation (Loh, Teixeira, & Lee, 2004) (see Table 1):

$$\Delta G = -RT \ \ln\left(\frac{\text{cac}}{\text{cmc}}\right) \tag{2}$$

where cac is the critical aggregation concentration.

The values of excess Gibbs energy are positive. The incorporation of surfactant molecules into the polymer-bound aggregates is energetically favorable.

The presence of a hydrophobic component in a polymer affects also the surface tension of aqueous solution (see Fig. 7). ι-Carrageenan does not affect the surface tension of water, while CAR-*graft*-PNIPAM lowers it considerably, from the value of 72 mN/m, characteristic of water, to 66 mN/m, at 25 °C. The effect of a surfactant on the surface tension of the polymer solutions was then studied. For CAR-*graft*-PNIPAM, even at a very low concentration of DTAC equal to $3.2{\cdot}10^{-7}$ M, a decrease of surface tension to the value of about 63 mN/m is observed. Then, the value of γ does not change in quite a wide range of DTAC concentrations, i.e. $3.2{\cdot}10^{-7}$–10^{-5} M. However, further increase of the surfactant concentration resulted in a rapid decrease of γ. Neither CAR-*graft*-PNIPAM nor DTAC (in this concentration range) alone do not show such effect. This observation suggests that in the CAR-*graft*-PNIPAM – DTAC system the surface active polymer–surfactant complexes are formed. Their surface activity increases with an increase of the DTAC content in a solution. For the ι-carrageenan – DTAC system the surface tension decrease starts at DTAC concentration around $3.2{\cdot}10^{-5}$ M. It decreases sharply and the dependence of γ vs. DTAC concentration reaches plateau at $3.2{\cdot}10^{-4}$ M. This plateau indicates that the polymer chain is saturated with the surfactant molecules. There is no such a saturation effect for the CAR-*graft*-PNIPAMI – DTAC system even at a much higher DTAC concentration ($\sim 10^{-2}$ M). This indicates that adsorption capability of the grafted polymer is much higher than that of the non-modified ι-carrageenan.

Comparative analysis of the changes in I_3/I_1 and surface tension upon increase of [DTAC] in CAR-*graft*-PNIPAMI allows for the following observations: The decrease in surface tension for non-modified carrageenan coincide with significant increase in I_3/I_1 while for CAR-*graft*-PNIPAMI surface tension the decrease starts at

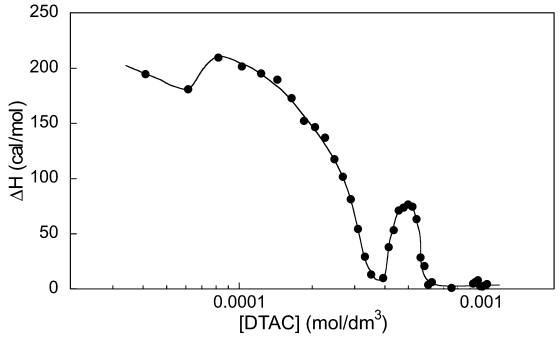


Fig. 8. Isothermal titration calorimetry thermogram for CAR–DTAC system, c_p = 0.1 g/L.

the [DTAC] two orders of magnitude lower than the increase of I_3/I_1. This suggests that, in contrast to non-modified carrageenan, surfactant binding to the CAR-*graft*-PNIPAMI polymer starts well before the aggregates appear. This reflects the difference in the nature between the surfactant interactions with non-modified polymers and the grafted ones. While binding of DTAC to ι-carrageenan is cooperative, the adsorption of the surfactant on CAR-*graft*-PNIPAMI does not lead to the formation of surfactant aggregates at the onset of binding. The behavior of CAR-*graft*-PNIPAMI – DTAC system resembles that observed for hydrophobically modified polyions interacting with oppositely charged surfactants. In such systems polymer hydrophobic domains have a capacity to solubilize individual hydrophobic molecules. Thus, single surfactant molecules may bind to hydrophobically modified polymer (Piculell, Guillemet, Thuresson, Shubin, & Ericsson, 1996). The results presented here are consistent with other reports that show that the binding of a surfactant to the hydrophobically modified polyion is stronger (takes place at lower surfactant concentrations), however, it is less cooperative (Benrraou, Zana, Varoqui, & Pefferkorn, 1992).

Based on the experimental data presented above one can suggest that the driving forces for the polymer – surfactant aggregation process differ considerably for the non-modified and grafted ι-carrageenan. Electrostatic interactions are expected to play the most important role in the system of ι-carrageenan – DTAC, while for the CAR-*graft*-PNIPAM – DTAC system the hydrophobic effect should be considered. To prove that assumption the isothermal titration calorimetry thermograms for the ι-carrageenan – DTAC and CAR-*graft*-PNIPAMI – DTAC systems were obtained (see Fig. 8).

As expected, the aggregation process of the anionic polysaccharide with the cationic surfactant was found to be endothermic and the thermogram shows two peaks. The presence of these two peaks in the enthalpy curve is characteristic of the systems composed of an oppositely charged polymer and surfactant (Heskins & Guillet, 1968) and indicates that DTAC interacts with ι-carrageenan in two stages. The first pronounced endothermic peak is believed to correspond to the electrostatic binding of the cationic headgroups of the surfactant molecules to the negatively charged sulphate groups along the polymer (ι-carrageenan) chains. It was shown that this electrostatic binding of the surfactant to the oppositely charged polymer and micellization are driven by entropy. The positive entropy for the electrostatic binding is attributed to the recovery of translational entropy of released counterions by the bound surfactant, whereas the entropy gain for micellization is attributed to the disruption of the water structure (Wang & Tam, 2002). The heat is absorbed at the first injection of the surfactant and further addition just increases that value. This indicates that the adsorption process is a cooperative phenomenon. According to the literature (Campo et al., 2009) the second, less pronounced peak, should be ascribed to the micellization of the surfactant molecules bound on the polymer chains.

For the CAR-*graft*-PNIPAM – DTAC system, in the same range of the surfactant concentrations, the enthalpy effect was below the detection limit. However, the surface tension measurements indicated clearly that there are some interactions between CAR-*graft*-PNIPAM and DTAC. The lack of measurable heat effect (evolved or absorbed) accompanying these interactions confirms that the driving forces for the aggregation occurring in that system are entropy driven. Most probably the aggregation occurs due to the so called "hydrophobic effect".

It should be pointed out that increasing polymer concentration, surfactant concentration, ionic strength, or temperature at the temperature range above LCST resulted in the increased turbidity of the solution and, at some point, in the phase separation, which is a typical behavior of the oppositely charged polymer–surfactant systems. This is important from the practical point of view since it allows using CAR-*graft*-PNIPAM for the removal (and thus purification) of cationic surfactants from water.

Conclusions

Novel thermosensitive polyelectrolytes were obtained by grafting PNIPAM onto ι-carrageenan. It was observed that in spite of a low degree of grafting the polymer is highly thermosensitive, with the LCST close to that characteristic of PNIPAM homopolymer. The LCST can be additionally adjusted by changing the ionic strength of the aqueous solution.

The interactions of ι-carrageenan and CAR-*graft*-PNIPAM with a model cationic surfactant – dodecyltrimethyl ammonium chloride were studied using three complementary physicochemical techniques: fluorescence molecular probe technique, surface tension measurements, and isothermal titration calorimetry. Critical aggregation concentration (cac) of a surfactant in the system containing the non-modified ι-carrageenan was lower than that for the grafted polymer. CAR-*graft*-PNIPAM was found to be surface active while the non-modified ι-carrageenan did not demonstrate this property. Complexes of DTAC with the grafted polysaccharide were also surface active even at very low surfactant concentrations. Results of the isothermal titration calorimetry suggest that the driving forces of the process of the ι-carrageenan – DTAC complexation were electrostatic. In the case of grafted polymer the driving forces for the complexation process were found to be of different nature. Most likely the introduction of NIPAM grafts to the polymer chain made the "hydrophobic interactions" with surfactant molecules dominating over the electrostatic.

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