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Poly[*N*-(2-hydroxypropyl)methacrylamide-*block*-*n*-butyl acrylate] micelles in water/DMF mixed solvents

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Abstract

Three diblock copolymers of poly[*N*-(2-hydroxypropyl)methacrylamide] (poly(HPMA)) and poly(*n*-butyl acrylate) (poly(BA)) with varying lengths of blocks were prepared by atom transfer radical polymerization. All copolymers were found to be soluble in dimethylformamide (DMF) and poorly soluble or insoluble in water. In water and mixed DMF/H_2O solvents, the copolymers were dispersed in micellar form by controlled addition of water to DMF solutions of copolymers under continuous intensive stirring. The micellar solutions in water were prepared by dialysis of solutions in DMF/H_2O (95 vol% of H_2O) against water. Solution properties of diblock copolymers of poly(HPMA) and poly(BA) were studied using static and dynamic laser light scattering to characterize the behavior of the copolymers at the supramolecular level. The effects of preparation mode, organic solvent (DMF) and copolymer chemical composition on the formation of micelles were studied. While a slower mixing procedure was optimal for copolymers with short poly(HPMA) blocks, a faster mixing was more suitable for copolymers having longer poly(HPMA) blocks. Finally, the dimensions of micelles in water were evaluated. The most compact micelles were prepared from copolymers having short hydrophilic poly(HPMA) blocks. On the other hand, the copolymer with the longest poly(HPMA) block formed micelles with the smallest size and the lowest density. © 2002 Elsevier Science Ltd. All rights reserved.

Keywords: Micellization; Poly[*N*-(2-hydroxypropyl)methacrylamide-*block*-*n*-butyl acrylate]; Dimethylformamide water mixed solvents

1. Introduction

Diblock copolymers associate and form micelles in selective solvents (i.e. in liquids which are good solvents for one block and precipitants for another) [1,2]. There have been tremendous advances over the past decade in the theory of block copolymer micelle formation [3–11]. At very low polymer concentrations, chains remain free in order to maximize their translational entropy. As the concentration is increased, one reaches a critical micellar concentration (CMC) where it is favorable for the insoluble blocks to associate and trade their translational freedom in order to reduce their enthalpy. The CMC is strongly dependent on the degree of incompatibility, χN_A(7); N_A is the length of the insoluble block and χ the quasibinary Flory–Huggins interaction parameter. For long chain diblock copolymers in selective solvents, it is well established that micellization occurs at relatively low concentrations and low values of χ [1,12–16]. Experimental evidence indicates that at concentrations close to the CMC, the micelles are spherical in shape; the block asymmetry determines the micellar geometry. At higher concentrations the number density of micelles increases, and eventually, entanglements between dangling blocks of the close packed micelles lead to ‘gel’ formation [17–19].

Recently, an interest in the formation of water soluble micelles from hydrophobic/hydrophilic block copolymers having hydrophobic cores and hydrophilic shells has rapidly increased [20–30]. Such studies are of direct relevance to many biomedical problems, including the controlled release of drugs in cancer tissue [31–36] and industrial applications in the removal or delivery of hydrophobic substances from aqueous media [8,37]. Kataoka et al. have been systematically evaluating block copolymers of poly(ethylene oxide) as the hydrophilic block and poly(aspartic acid) blocks with

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Table 1
Characteristics of copolymers

Sample	M_n	M_w/M_n	PBA block M_n	HPMA/BA[a] molar ratio	HPMA molar fraction	$(dn/dc)_{H_2O}$ (ml g^{-1})
1	23 100	1.42	13 200	0.67	0.40	0.144
2	34 000	1.69	9900	2.18	0.68	0.160
3	12 200	1.92	2000	4.56	0.82	0.182

[a] Calculated from NMR data.

doxorubicin (anticancer drug) covalently bound to carboxyl groups of aspartic acid residues as the hydrophobic block [31–33]. Such copolymers form micelles with the hydrophobic drug-binding segment as the core and the poly(ethylene oxide) block as the hydrophilic corona. Similar micelles from poly(ethylene oxide)-*block*-poly(β-benzyl-L-aspartate) were suitable for entrapment and release of doxorubicin [35].

Here, we have investigated the behavior of amphiphilic diblock copolymers poly[*N*-(2-hydroxypropyl)methacrylamide-*block*-*n*-butyl acrylate] (poly(HPMA-*b*-BA)), synthesized by atom transfer radical polymerization (ATRP) [38,39], in aqueous and mixed dimethylformamide (DMF)/water solutions and determined the size and molecular weight of the micelles by static and dynamic light scattering techniques.

2. Experimental part

2.1. Preparation of copolymers

The block copolymers were prepared by ATRP [38–40].

The poly(*n*-butyl acrylate) with a bromine end group (PBA–Br), prepared by ATRP [41,42] was introduced into a round bottom flask capped with a rubber septum and cycled three times between vacuum and nitrogen to remove oxygen. The degassed ethanol was added via a syringe, and the mixture (20 wt%) was stirred until a homogeneous solution was formed. Separately, in another flask, the calculated amounts of 1,4,8,11-tetramethyl-1,4,8,-11-tetrazacyclotetradecane (Me4Cyclam), CuBr, $CuBr_2$ and HPMA were introduced (PBA–Br/CuBr/$CuBr_2$/Me4Cyclam = 1:1:0.2:1.2) [43]. The amount of HPMA depended on the target M_n of the poly(HPMA) block [44]. The flask was capped with a rubber septum and cycled three times between vacuum and nitrogen. The macroinitiator solution was then transferred under nitrogen, via canulla, into the flask containing the monomer and the initiating system. The flask was then placed into an oil bath thermostated at 50 °C. After 18 h, the reaction mixture was cooled down, the flask opened and the content diluted with ethanol. Cation exchange resins were added and the reaction mixture was stirred for 1 h [45]. The resin was removed by filtration and the yellowish polymer solution was subjected to dialysis to remove unreacted monomer. Finally, the block polymer was recovered by removing the solvent under vacuum.

Molecular characteristics of diblock copolymers are summarized in Table 1 and their structures are shown in Fig. 1.

2.2. Solvents

Deionized water (Q-water) and distilled DMF (Fluka) were used for light scattering experiments.

2.3. Preparation of micelles

The solubility of poly(HPMA-*b*-BA) copolymers in water was found to be a function of the molar content of hydrophilic HPMA. While copolymers **1** and **2** with lower contents of poly(HPMA) were not dispersible in water, copolymer **3** was soluble in water, forming large pseudomicelles. Nevertheless, all the copolymers were found to be molecularly soluble in DMF at room temperature. A special procedure had to be used to disperse them in the micellar form in water and mixed DMF/H_2O solvents with a higher volume content of water. These copolymer solutions were prepared directly in measuring cells with a diameter of 18 mm by the controlled addition of water to DMF solutions of copolymers under continuous intensive stirring (400 rpm). The magnetic stirrers with the length of 10 mm and diameter of 3.5 mm were used. The concentration of the final solution was always $c = 0.001$ g ml^{-1} and the final sample volume was 4 ml. This method is somewhat analogous to the traditional stepwise dialysis procedures introduced in Refs. [24,46] the method of a regulated addition of

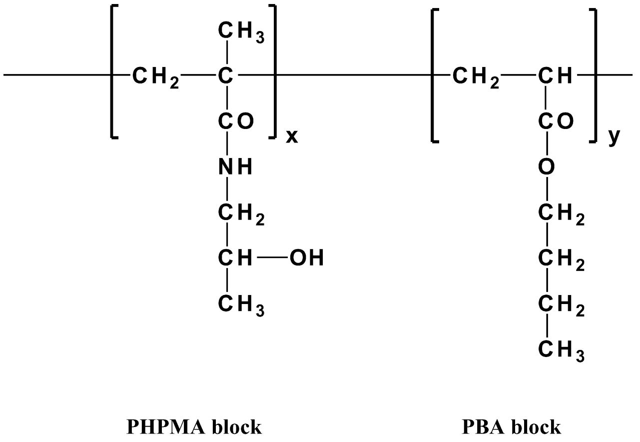

Fig. 1. Structure of poly[*N*-(2-hydroxypropyl)methacrylamide-*block*-*n*-butyl acrylate].

selective solvent applied by the group of Eisenberg [47,48] but allows for better control of the speed of preparation. A syringe pump was used to control the speed of water addition. The preparation time, t_p, was varied from 0.333 to 278 min. The mixing rates yielding copolymer solutions with the highest micellar and the lowest aggregate contents were selected for further processing. If the preparation parameters were fixed, then the reproducibility of the weight-average molecular weight of micelles was about 30%. The micellar solutions in water were prepared by dialysis of solutions in DMF/H_2O (95 vol% of H_2O) against water. The dialysis membrane Spectra/Por 1 (Spectrum, USA) manufactured from regenerated cellulose with a molecular weight cut off of 6000–8000 were used. The solutions for light scattering measurements were optically purified by filtration with filters having a pore size of 0.45 or 0.8 μm.

2.4. Static light scattering

Static light scattering measurements were performed with a commercial Sofica instrument in vertically polarized light of a He–Ne laser at wavelength $\lambda_0 = 632.8$ nm, angular range $\theta = 30$–150°, and temperature 25 °C. The apparatus was calibrated with benzene as a standard. The processed data are represented (unless otherwise noted) as

$$Kc/R(\theta, c) = (M_w P(\theta))^{-1} + 2A_2 c, \tag{1}$$

where M_w is the weight-average molecular weight; K the optical constant which includes the square of the refractive index increment dn/dc; $R(\theta)$ the Rayleigh ratio, proportional to the intensity of light scattered from solutions, A_2 the second virial coefficient, and c the (co)polymer concentration in g ml^{-1}. $P(\theta)$ is the particle scattering function. M_w can be evaluated from the zero angle and concentration limit of $Kc/R(\theta, c)$. If the concentration dependence of $Kc/R(\theta, c)$ was not available and $A_2 \neq 0$, only the apparent values of the weight-average molecular weight, $M_w^a (= Kc/R(\theta, c))^{-1}$ were estimated.

Since the copolymers are not directly soluble in water, the refractive index increments, dn/dc, of copolymers could not be directly measured. Therefore, we measured the increments with the Brice–Phoenix differential refractometer at $\lambda = 630$ nm in DMF, as all copolymers were well soluble. The $(dn/dc)_{H_2O}$ values of copolymers in water (Table 1) were estimated from values $(dn/dc)_{DMF}$ in DMF by considering the actual difference in the refractive indexes of solvents.

2.5. Dynamic light scattering

Polarized dynamic light scattering (DLS) measurements were made in the angular range 30–135° using a light scattering apparatus equipped with an He–Ne (632.8 nm) and an Ar-ion laser (514.5 nm) and an ALV 5000, multi-bit, multi-tau autocorrelator covering approximately 10 decades in a delay time t. Most of the measurements were made at the scattering angle $\theta = 90°$.

The inverse Laplace transform using the REPES [49] method of constrained regularization (which is similar in many respects to the inversion routine CONTIN [50]) was used for an analysis of autocorrelation functions. REPES directly minimizes the sum of the squared differences between the experimental and calculated intensity time correlation functions using nonlinear programming. This method uses an equidistant logarithmic grid with fixed components (here a grid 10 components per decade) and determines their amplitudes. As a result, a distribution function $A(\tau)$ of decay times is obtained. From the characteristic decay times, τ_i (the peak positions of $A(\tau)$) the corresponding apparent average diffusion coefficient, $D_i^a(90°)$, was calculated from the equation:

$$D_i^a(90°) = 1/\tau_i q^2, \tag{2}$$

where q is the absolute value of the scattering vector. The apparent average hydrodynamic radius, R_{hi}^a, was calculated from $D_i^a(90°)$ using the Stokes–Einstein equation:

$$R_{hi}^a = kT/6\pi\eta D_i^a(90°), \tag{3}$$

where k is the Boltzmann constant, T absolute temperature and η the viscosity of solvents at 25 °C (Table 1). The hydrodynamic radius, R_{hi}, was calculated from the zero angle and concentration limits of D_i^a. The viscosities of DMF/water mixed solvents were determined with a capillary viscometer; values are shown together with corresponding solvent densities in Table 2.

The experimental error of the R_{hi}^a-determination for micelles was typically about 3%.

2.6. Evaluation of the weight-average molecular weight of micelles in presence of aggregates

In the presence of aggregates in solutions, M_{wm} of micelles were estimated using a combination of dynamic and static light scattering experiments. If two or more kinds of particles are present in solution, the total scattered intensities, $I_{st}(\theta)$, can be expressed as a sum of individual

Table 2
Viscosities and densities of mixed solvents DMF/water

Vol% of H_2O	η (mPa s)	ρ (g ml^{-1})
0	0.810	0.9445
10	1.330	0.9648
20	1.950	0.9790
30	2.392	0.9889
40	2.502	0.9951
50	2.312	0.9976
60	2.010	0.9986
70	1.688	0.9982
80	1.389	0.9979
90	1.127	0.9975
95	1.085	0.9981
100	0.894	0.9985

contributions. In the case of a mixture of micelles and aggregates

$$I_{\mathrm{st}}(\theta) = I_{\mathrm{sm}}(\theta) + I_{\mathrm{sa}}(\theta), \tag{4}$$

where $I_{\mathrm{sm}}(\theta)$ and $I_{\mathrm{sa}}(\theta)$ are the total scattering intensities generated by micelles and aggregates, respectively. In addition to the hydrodynamic size of micelles (R_{hm}) and aggregates (R_{ha}), the analysis of dynamic light scattering data also provides us with the relative scattering amplitude $f_{\mathrm{ra}}(\theta) = I_{\mathrm{sa}}(\theta)/I_{\mathrm{st}}(\theta)$. Therefore Eq. (4) can be rewritten:

$$I_{\mathrm{sm}}(\theta) = I_{\mathrm{st}}(\theta) - I_{\mathrm{sa}}(\theta) = I_{\mathrm{st}}(\theta)(1 - fr_{\mathrm{a}}(\theta)). \tag{5}$$

Thus, the scattered intensity $I_{\mathrm{sm}}(\theta)$ generated only by micelles can be extracted from the total scattered intensities $I_{\mathrm{st}}(\theta)$. The procedure can be successfully used when $I_{\mathrm{sm}}(\theta)$ is comparable with $I_{\mathrm{sa}}(\theta)$ (a small amount of aggregates) and the distribution of aggregate sizes is sufficiently distinct from the average micelle size. Since both the assumptions are fulfilled in systems under investigation (Section 3) $M^{\mathrm{a}}_{\mathrm{wm}}$ of micelles can be estimated from Eq. (1) using $I_{\mathrm{sm}}(\theta)$ values and the concentration of micelles, c_{m}, instead of c. The concentration c_{m} is generally unknown but in particular case of here investigated copolymer solutions the mass fraction of aggregates is small. If we assume that the same hard-sphere model is applicable for both types of particles (M_{w} of particles is proportional to R_{h}^3), then the mass fraction of aggregates for $I_{\mathrm{sm}}(\theta) \sim I_{\mathrm{sa}}(\theta)$ is in the order of 0.01. M_{wm} was then calculated from the zero concentration limits of $M^{\mathrm{a}}_{\mathrm{wm}}$.

3. Results and discussion

Solution properties of diblock copolymers of poly(HPMA) and poly(BA) were evaluated using static and dynamic laser light scattering to characterize the behavior of the copolymers at the supramolecular level. The effect of preparation rate, composition of mixed DMF/H_2O solvent and copolymer composition on the formation of micelles of poly(HPMA-*b*-BA) copolymers were studied. Finally, the particle parameters of micelles in water were evaluated.

3.1. Effect of preparation rate on association of copolymers in mixed DMF/H_2O solvents with 95 vol% of H_2O

The effect of the preparation time of copolymer solutions, t_{p}, on association of copolymers was investigated by the dynamic light scattering spectroscopy in mixed DMF/H_2O solvents with 95 vol% of H_2O. Two populations of particles were generally observed in the solutions. The small particles with a narrow distribution of sizes were assigned as micelles while the polydisperse large particles are probably aggregates of the copolymers. The apparent hydrodynamic radius of micelles, $R^{\mathrm{a}}_{\mathrm{hm}}$, and aggregates, $R^{\mathrm{a}}_{\mathrm{ha}}$, and the relative scattering amplitude of micelles, f_{rm}, are plotted as a function of t_{p} for copolymers **1** and **2** in Figs. 2 and 3. The concentration of final solutions was always $c = 0.001\ \mathrm{g\ ml^{-1}}$ and the composition of the final mixed solvent was 95 vol% of water and 5 vol% of DMF. Comparison of Figs. 2 and 3, shows that the optimal t_{p} defined by the lowest aggregate content was strongly influenced by molar fraction of HPMA. While for copolymer **1** with a 0.40 M fraction of HPMA a slower mixing procedure was optimal, faster mixing was better for the copolymer having the 0.68 M fraction of HPMA. For further processing, t_{p} of 17 and 2.78 min were selected for copolymers **1** and **2**,

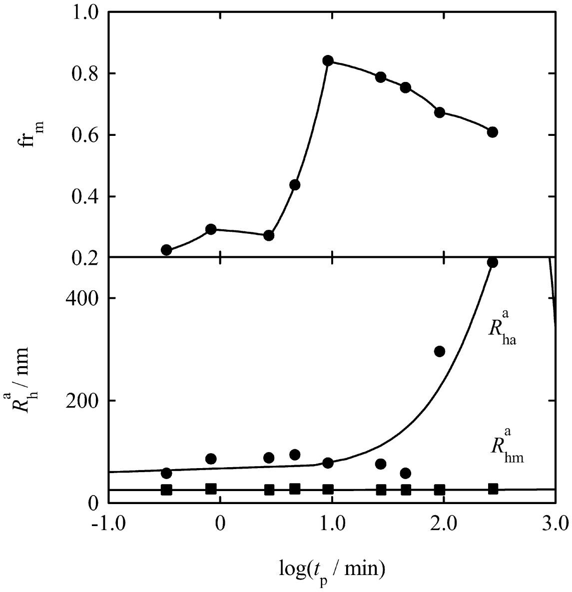

Fig. 2. Dependence of the apparent hydrodynamic radius of micelles, $R^{\mathrm{a}}_{\mathrm{hm}}$ and aggregates, $R^{\mathrm{a}}_{\mathrm{ha}}$, and the fraction of scattered light of micelles in solution, f_{rm}, on preparation time, t_{p}, for copolymer **1**.

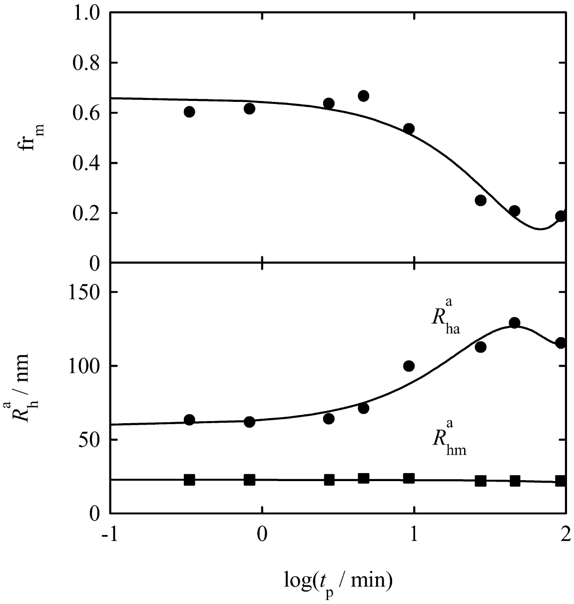

Fig. 3. Dependence of the apparent hydrodynamic radius of micelles, $R^{\mathrm{a}}_{\mathrm{hm}}$ and aggregates, $R^{\mathrm{a}}_{\mathrm{ha}}$, and the fraction of scattered light of micelles in solution, f_{rm}, on preparation time, t_{p}, for copolymer **2**.

respectively. In spite of the fact that the scattering intensities of micelles and aggregates were comparable, the weight fraction of the aggregates was small in the order of 0.01. Thus, relatively good micellar solutions suitable for biomedical and industrial applications can be prepared by this procedure.

Copolymer **3** with the high content of HPMA (molar fraction of HPMA = 0.82) was well soluble in the mixed DMF/H_2O solvent with 95 vol% of water. No effect of preparation rate was observed in the time window used. Moreover, the copolymer **3** could be directly dispersed in water. Unfortunately, the particles obtained by direct dissolution in water were larger than those prepared by dialysis from solutions in DMF/H_2O revealing an insufficient solubility of the copolymer in water. $M_w^a = 1.8 \times 10^7$ g mol^{-1} and $R_h^a = 55$ nm were found for these particles (compare with data for copolymer **3** in Table 3).

3.2. Association of copolymers in mixed solvents of DMF and water

The earlier experimental observations gave rise to a question of how the micelles and copolymer aggregates are formed during the preparation. Therefore, we started a detailed DLS investigation of the solution properties of the copolymers in DMF/H_2O mixtures. The rate of mixing for copolymer **1** and **2** was selected to be the same as in Section 3.1 and the final concentration of copolymers was always $c = 0.001$ g ml^{-1}. The micellar solutions in water were prepared by dialysis of solutions in DMF/H_2O (95 vol% of H_2O) against water. The results of DLS measurements are shown in Fig. 4. Fig. 4 shows that diverse association procedures take place as a function of the relative length of poly (HPMA) blocks. Thus, large polydisperse aggregates with $R_{ha}^a \geq 100$ nm were formed at low H_2O volume fractions and micelles were visible at H_2O volume fractions higher than 60 vol% for copolymer **1**. The relative scattering amplitude of micelles, f_{rm}, increased with increasing water content (see insert in Fig. 4(a)). The opposite situation was observed in solutions of copolymer **2** (Fig. 4(b)). Micelles without aggregates were already formed at 20 vol% of H_2O in the solvent and R_h^a was practically independent of water content. Formation of aggregates was observed only at high water content in the solvents ($f_{ra} = 0.34$ and 0.46 was found for 95 and 100 vol% of H_2O). In the case of copolymer **3**, a successive increase of both R_h^a was observed with increasing H_2O content (Fig. 4(c)). The most compact micelles were observed in aqueous solutions.

The earlier results show that the random association of copolymers dominates in the self-association of copolymers with short poly(HPMA) blocks (**1**) at low volume fractions of water. The process of self-association is successively changed with increasing water content to micellization at higher water contents. The change of association processes is probably induced by increasing differences in solubility of poly(HPMA) and poly(BA) blocks in mixed solvents due to an increase in water content. We have previously observed similar behaviors in solutions of graft copolymers [51,52]. Thus, a low difference in solubility of the blocks is sufficient for random association, while a higher one is necessary for micellization. The self-association process in solutions of copolymers with longer poly(HPMA) blocks is dominated by micellization due to a higher 'polar' character of the copolymers (due to an increased ratio of hydrophilic HPMA blocks) already at low water contents in accordance

Table 3
Characteristics of micelles in aqueous solutions (M_{wm}: weight-average molecular-weight of micelles in aqueous solutions, R_{hm}: *z*-average hydrodynamic radius of micelles in aqueous solutions, w_m: weight fraction of copolymers in the micelle)

Sample	$M_{wm} \times 10^6$ (g mol^{-1})	R_{hm} (nm)	w_m (g ml^{-1})
1	19.4[a]	28.8[a]	0.32
2	8.3	23.8	0.25
3	1.3	13.4	0.21

[a] Apparent values in water at $c = 0.001$ g ml^{-1}.

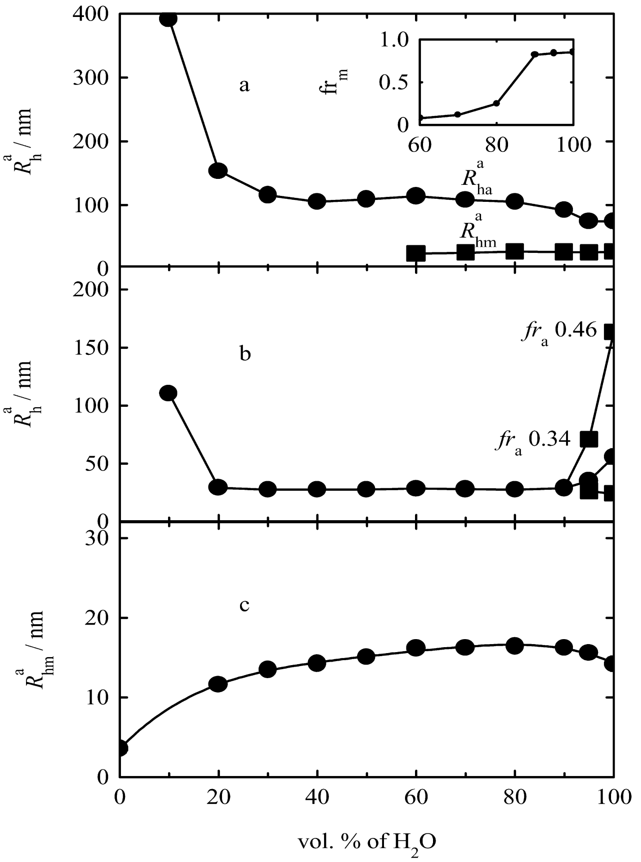

Fig. 4. (a) The hydrodynamic radius of micelles, R_{hm}^a, and aggregates, R_{ha}^a, of copolymer **1** in DMF/H_2O mixed solvents plotted as a function of water content. The fraction, f_{rm}, is shown in the insert. The preparation time was 17 min. (b) The same as in (a) for copolymer **2**. The hydrodynamic radius of micelles, R_{hm}^a, and aggregates, R_{ha}^a, are shown for solvents with 95 and 100 vol% of water by filled squares. Corresponding values of f_{ra} are given in the annotations. The preparation time was 2.78 min. (c) The same as in (a) for copolymer **3**.

with previous data [52]. For the relatively well water soluble copolymer **3**, the mean weight fraction of copolymer in micelles qualitatively follows a decrease in the thermodynamic quality of the solvents for poly(BA).

3.3. Micellar parameters in aqueous solutions

The micellar solutions in water were prepared by dialysis of solutions in DMF/H_2O (95 vol% of H_2O, $c = 0.001$ g ml^{-1}) against water and then diluted as necessary. In order to estimate the molecular weight M_{wm} and hydrodynamic radius R_{hm} of the micelles, the concentration of dependence of M^a_{wm} and R^a_{hm} were measured and extrapolated to $c = 0$. To eliminate the contribution of aggregates to light scattering intensity the combination of static and dynamic light scattering measurements was used. Applicability of the procedure is supported by the REPES analysis in Fig. 5. R_h-distributions of micelles and aggregates are well distinguished and the scattering intensities of micelles and aggregates are comparable. The concentration dependencies of M^a_{wm} and R^a_{hm} for copolymers **2** and **3** are shown in Fig. 6. Both the parameters did not show any upward curvature in the region of low concentrations, typical of micelle dissociation. This means that the association equilibrium was strongly shifted in favor of micelles within the whole range of measured concentrations. Moreover, the second virial coefficients were low, as generally observed for other micellar solutions [1,53]. The characteristics of micelles in aqueous solutions are listed in Table 3. The mean weight fractions, w_m, of copolymer in the micelles (copolymer density) were calculated from the volumes of micelles V_m (in cm^3) and from their corresponding molecular weights M_{wm} (in g mol^{-1}); $w_m = M_{wm}/N_A V_m$, where N_A is Avogadro's number. Since micelles can be well approximated with a model of solid spheres [54], R_{hm} values were used for the estimation of V_m. From Table 3 it can be seen that the most compact micelles are prepared from copolymers having a low content of hydrophilic poly(HPMA). It is probably due to strong attractive hydrophobic interactions of poly(BA) chains and a low solvation effect of the short poly(HPMA) blocks. On the other hand, the copolymers with high content of poly(HPMA) formed micelles of small sizes with low densities, which reflects an overall high solubilization effect of long poly(HPMA) chains. The weight fractions of the studied copolymers, w_m, were comparable to those observed with micellar solutions in selective organic solvents [53]. The micelles of copolymer **3** prepared by dialysis were smaller than particles prepared by directly dissolving the copolymer in water. It is probably due to a low solubility of the copolymer in water; the ordered structure of copolymers already existing in the solid state could not be destroyed and rearranged to the equilibrium micellar form. We have observed a similar behavior in decane and dioxane solutions of poly(styrene-*block*-hydrogenated polybutadiene) copolymers [55].

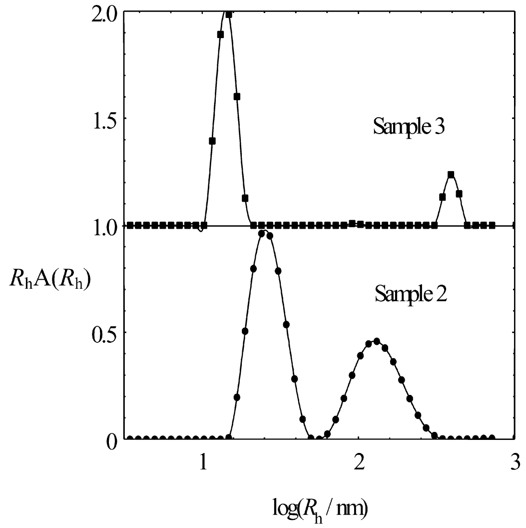

Fig. 5. R_h-distributions of micelles and aggregates in aqueous solutions of copolymer **2** and **3** ($c = 0.001$ g ml^{-1}) as obtained by the REPES analysis of DLS data measured at $\theta = 90°$.

4. Conclusion

The poly(HPMA-*b*-BA) diblock copolymers can be dispersed in water by the controlled addition of water to DMF solutions of copolymers under continuous intensive stirring and subsequent dialysis of solutions in DMF/H_2O (95 vol% of H_2O and 5 vol% of DMF) against water in wide range of copolymer compositions (the molar fraction of

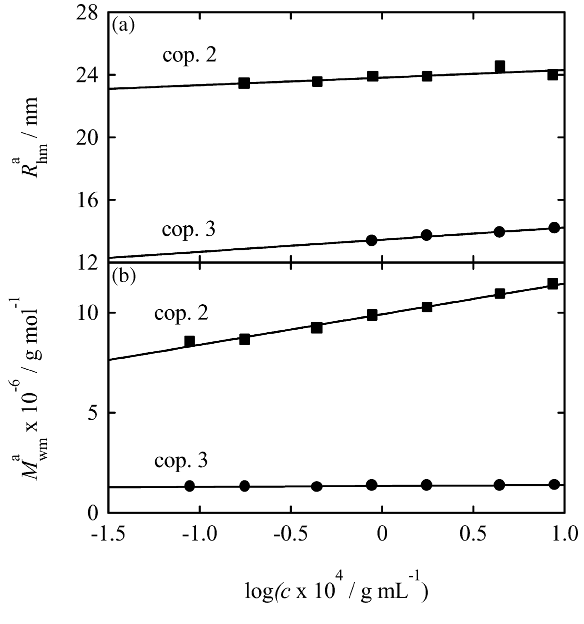

Fig. 6. Concentration dependence of the weight-average molecular weight, M^a_{wm} (a) and the *z*-average hydrodynamic radius, R^a_{hm}, of micelles in water (b) for copolymer **2** and **3**. The micellar solutions were prepared by appropriate dilution of solutions of the copolymers prepared by dialysis of DMF/H_2O solutions (solvent composition: 95 vol% of water and 5 vol% of DMF) with $c = 0.001$ g ml^{-1} against water.

HPMA from 0.40 to 0.82). While a slower mixing procedure was optimal for copolymers with short poly(HPMA) blocks, faster mixing was better for copolymers having longer poly(HPMA) blocks. The most compact micelles were prepared from copolymers having only short hydrophilic poly(HPMA) blocks. On the other hand, copolymers with longer poly(HPMA) blocks formed micelles of smaller sizes with relatively lower densities.

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