Monodisperse Particles Based on Copolymers of Methyl methacrylate or Styrene with *N*-Vinylformamide

Anastasia Menshikova, Tatiana Evseeva, Natalya Shevchenko, Boris Shabsels, Alexander Yakimansky, Sergey Ivanchev*

Sammary: Various techniques of one-step batch emulsion copolymerization of methyl methacrylate (MMA) or styrene (St) with *N*-vinylformamide (NVF) initiated by 2,2′-azobis(2-methylpropanamidine) (AIBA) or 2,2′-azobis[4,5-dihydroimidazol-2-yl] (AIP) dihydrochlorides in the presence of dextran, cetyltrimethylammonium bromide (CTAB) or without any stabilizers were examined to obtain monodisperse submicron and nano- particles, having both positive surface charge and hydrophilic surface. After hydrolysis in acidic media, the particle surface contained amino groups originating from NVF units along with carboxylic groups from hydrolyzed initiator residues. The obtained particles were tested as building blocks of hierarchic structures. Owing to their amphiphilic surface, the particles were capable of self-assembling from their dispersions into three-dimentional (3D) ordered arrays.

Keywords: amphiphilic surface; emulsion copolymerization; monodisperse particles; *N*-vinylformamide; self-assembly

Introduction

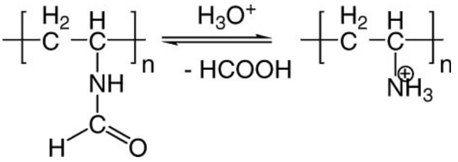
Monodisperse polymeric particles with functional groups on their surface are of great interest for their applications as carriers for biologically active substances[1] or as building blocks of novel hierarchic materials.[2,3] The particle size, surface structure, and functional group nature are the main factors, which should be regulated during heterophase polymerization to meet certain requirements in these fields.[3–5] However, the introduction of amino groups into particle surface layers causes some troubles, since these groups interfere with radical polymerization processes. Therefore, a two-step procedure was proposed by Pichot and coauthors.[6] First, poly(vinylbenzyl chloride) latexes were prepared in emulsifier-free conditions using 2,2′-azobis(2-methylpropanamidine) (AIBA) as an initiator; then the resulting latexes were reacted with trimethylamine. Next, batch emulsion copolymerization of vinylbenzylamine hydrochloride (VBAH), styrene and divinylbenzene (DVB) using AIBA was examined,[7] but secondary nucleation could occur in the presence of the water-soluble functional comonomer. Okubo also copolymerized vinylbenzylchloride with styrene and DVB using polystyrene seeds and then modified the particle surface with polyamines.[8] Latexes containing (*tert*-butoxycarbonyl)amino (Boc) groups were prepared by dispersion copolymerization of styrene and Boc-aminostyrene (Boc-ASt) in isopropyl alcohol; after synthesis amino groups were deprotected by the treatment with 2 M HCl in isopropyl alcohol/water at 50 °C for several hours, resulting in $NH^{+}{}_{3}Cl^{-}$ groups on the particle surface.[9] Multistep batch emulsion polymerization was also applied to obtain amino groups on the particle surface.[10] In the first one or two steps, monodisperse cationic polystyr-

---

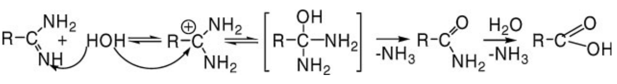
Institute of Macromolecular Compounds, Russian Academy of Sciences, Bolshoi pr. 31, St. Petersburg, 199004 Russia
Fax: +7(812)3286869; E-mail: asya@hq.macro.ru

ene seed particles were synthesized; then functional comonomers, (3-methacrylamidopropyl)trimethylammonium chloride, 2-aminoethyl methacrylate hydrochloride and VBAH, were used to synthesize the final latexes using quaternary ammonium emulsifiers and AIBA.

All the above mentioned methods are very complicated and cannot ensure both narrow particle size distribution (PSD) and high conversion of amino-containing water-soluble comonomers, in particular the cationic ones. *N*-vinylformamide as a functional comonomer could give a good chance to overcome the problem. In such a case, amino surface groups could appear due to surface hydrolysis of NVF units in acidic media after copolymerization:[11]



By this approach, core-shell particles were prepared using PS seeds and adding NVF in the last polymerization step.[12] However, in the case of batch styrene and NVF copolymerization, NVF mainly polymerized in solution and latexes lost their aggregate stability during the synthesis. It was proposed that water-soluble PNVF was not very susceptible to hydrogen abstraction, which is necessary to form polystyrene grafted-PVNF. Besides, due to protonation of the amino groups as well as terminal amidine groups of AIBA residues, the surface of these particles is assumed to be positively charged in a wide pH range. However, the presence of carboxylic groups on the particle surface, whose ionization provided negative surface charge at $pH > 10$, was observed. These carboxylic groups can be formed by hydrolysis of AIBA residues as was shown earlier:[13,14]



In our opinion, destabilization of latexes synthesized with AIBA was mainly caused by this side process. Therefore in our previous work,[15] AIP having imidazoline rings less prone to hydrolysis was used as an initiator in semicontinuous emulsifier-free copolymerization of styrene and NVF in aqueous media with addition of Boc-ASt or ASt hydrochloride in the last polymerization step. This approach was successfully applied to prepare monodisperse core-shell latex particles with surface amino groups. However, the preparation of amino-functionalized monodisperse latexes by one-step emulsion polymerization has been a challenging task so far.

The other way to stabilize latex particles during polymerization is the addition of polymer stabilizers and emulsifiers. For instance, dextran, capable of hydrogen abstraction and grafting in radical processes, was studied in our previous works as a steric stabilizer in MMA polymerization acting with 4,4′-azobis(4-cyanoisovaleric acid) in aqueous media.[16,17] Therefore, this work is devoted to examination of one-step batch styrene or MMA emulsion copolymerization with NVF, in the presence of dextran and CTAB or without any stabilizer, as a way to obtain monodisperse submicron and nanoparticles with surface amino groups.

Experimental Part

Styrene, MMA, NVF were purchased from Aldrich and distilled using standard techniques. Radical initiators, AIBA and AIP (Wako Pure Chemical Industries Ltd.), cetyltrimethylammonium bromide (CTAB) (Serva) and dextran (MW 60.000 ± 10.000), all of commercial grade, were used without additional purification.

Emulsion polymerization was carried out in a four-necked glass reactor with a glass paddle-type stirrer, a condenser, an argon inlet, and a thermostat. One half of NVF, the initiator, CTAB, and dextran were dissolved in double-distilled water refluxed and cooled in argon flow. Then the mixture of St or MMA with the second part of NVF was added and the initial pH value (pH_0) was adjusted to the weakly acidic range with aqueous HCl or NaOH. Continuous stirring at 300 rpm and a constant temperature in the range of 70–90 °C were maintained during the polymerization. After the synthesis, the residual monomers were removed by distillation.

Latex particle size was measured by transmission electron microscopy (JEOL JEM 100 S microscope, Japan). PSD was characterized by the root-mean-square deviation, σ. The surface concentration of weakly acidic and basic groups was determined by conductometric titration after three-fold washing of the latexes by successive centrifugation to remove water-soluble impurities followed by redispersion in double-distilled water. Latexes of nanoparticles were thoroughly dialyzed against double-distilled water. The synthesis conditions and characteristics of latexes are given in Table 1.

The electrophoretic mobility of the particles was studied using standard microelectrophoresis in 10^{-3} and 10^{-2} mol/L NaCl solutions over the pH range 2.0–10.9 and used for calculation of ζ-potential of the particles by Smoluchowski equation.

Thin films of the particles were fabricated from their aqueous dispersions by self-assembling on clean glass slides. The detailed techniques for film preparation are given elsewhere.[18] The structure of the thin films was examined by scanning electron microscopy (JEM 100 C JEOL microscope, Japan). For nondestructive monitoring of ordered structures in the films as well as in polymer dispersions, transition spectroscopy in the visible range was used.

Results and Discussion

Using one-step batch emulsion MMA or St copolymerization with NVF under conditions described in Table 1, we managed to prepare monodisperse particles of diameters in the range 75–375 nm with quite narrow PSD (Table 1, Figure 1). Copolymerization was carried out in slightly acidic

Table 1.
Copolymerization conditions and characteristics of latexes.

Latex									
	Comonomers w/w	T °C	Initiator[b] wt.%	CTAB mmol/L	Dextran[b] wt.%	pH_0	D nm	σ %	$NH_2 + COOH$[c] $\mu mol/m^2$
Submicron particles									
S1	MMA/NVF 100/5	65	AIBA 0.20	–	0.2	6.3	375	2.5	1.5/-
S2	MMA/NVF 100/10	65	AIP 0.20	–	0.2	6.0	350	3.8	0.5/-
S3	St/NVF 100/1.3	70	AIP 0.37	–	–	6.2	250	2.9	0.2/1.1
S4	St/NVF [d] 100/3	70	AIP 0.30	–	–	6.6	220	2.8	0.2/1.4
Nanoparticles									
N1	St/NVF 100/3	90	AIP 0.18	1.3	–	6.1	90	1.5	–
N2	MMA/NVF 100/3	70	AIP 0.18	0.9	0.2	6.2	75	1.9	–

[a] S1–S4: 10 wt.% comonomers per aqueous phase, N1 and N2: 5 wt.% comonomers per aqueous phase.
[b] Per aqueous phase.
[c] Without/after surface hydrolysis as a result of HCl/NaOH treatment.
[d] Mercaptoethanol was added into the reaction mixture – 0.2 wt.% per aqueous phase.

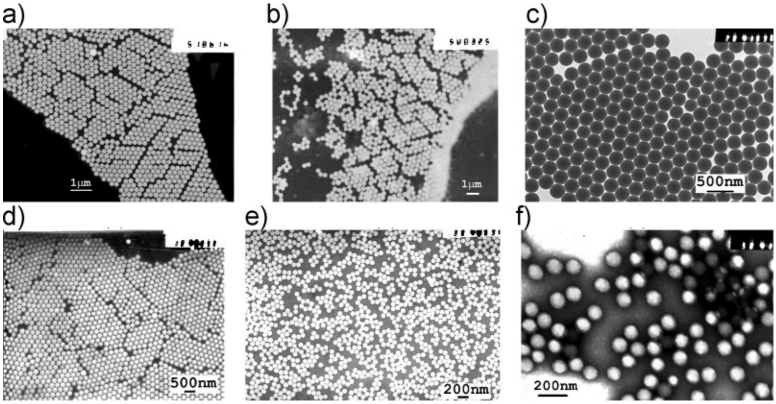


Figure 1.
TEM images of obtained latex particles. Latexes S1 (a), S2 (b), S3 (c), S4 (d), N1 (e), N2 (f) listed in Table 1.

media to prevent the hydrolysis of cationic groups of both radical initiators and their residues on the particle surface. However, in the presence of AIBA, in spite of the dextran-stabilizing ability, MMA and NVF copolymerization at above 5 wt.% NVF in comonomer mixtures led to coagulation during synthesis. Such behavior was caused by the hydrolysis of AIBA residues forming carboxylic groups on the particle surface. As a result, both the particle surface charge and latex stability decreased. Owing to the heterocyclic structure, the imidazoline groups are more stable.[13,14] Hence, we used AIP in our further experiments to provide sufficiently high positive surface charge of the latex particles during synthesis. This allowed us to decrease the coagulum formation. We found that 10 wt.% NVF relative to MMA and substitution of AIBA with AIP slightly decreased particle diameter going from S1 to S2 latex samples, which could be explained by enhanced positive surface charge in the nucleation stage.

Since certain differences were found in MMA and NVF copolymerization in the presence of AIBA and AIP, we studied how the initiators affect the stability of the surface characteristics of obtained particles during their storage and subsequent treatment. We found the surface charge of the resulting particles remained positive after storage for five months at 4 °C for the latexes prepared in the presence of both initiators. However, pH effects on ζ-potential values display an inversion of the ζ-potential sign (Figure 2). This indicates the presence of surface carboxylic groups when using AIBA (Figure 2a).

After one-day storage of latexes in acidic and alkaline solutions, their positive charge was retained in wide pH ranges owing to hydrolysis of NVF units on the particle surface forming additional amino groups along with carboxylic ones. S2 latexes

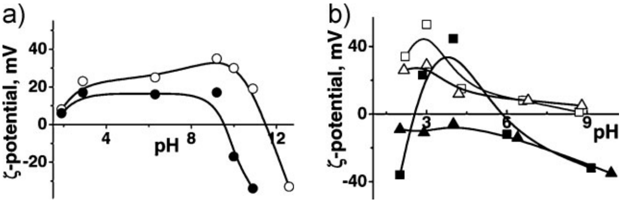
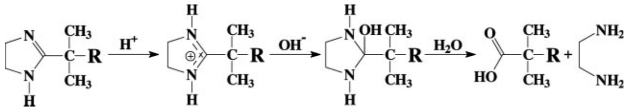


Figure 2.
ζ-potential of S1 (a) and S2 (b) latex particles (10^{-3} mol/L NaCl); ○, □, △ - immediately, ●, ■, ▲ - after one day storage in solutions of given pH; latex S2 □, ■ - without and △, ▲ - after HCl/NaOH treatment.

prepared with AIP were also thoroughly hydrolyzed after synthesis in the presence of 0.1 M HCl at 50 °C for 24 h and then washed with 0.01 NaOH solution and subsequently with double-distilled water. This treatment seemed to hydrophilize particle surface due to enrichment in both carboxylic and amino groups, although the negative sign of ζ-potentials prevailed. Thus, the applied HCl/NaOH treatment could cause even hydrolysis of AIP terminal residues:



Emulsifier-free styrene and NVF emulsion copolymerization was carried out without any stabilizers using lower NVF and higher AIP contents. In the process, NVF-enriched polymer chains were formed in the initial stage and took part in the nucleation. Localized at the particle surface, these chains formed a hydrophilic layer, which ensured particle aggregate stability due to both steric and electrostatic repulsion. The method ensured formation of monodisperse particles, but the conversion in S3 synthesis was not quite efficient – only 51 wt%. To promote fast nucleation at the initial stage and NVF copolymerization in the growing particles, mercaptoethanol as a chain transfer agent was added to reaction mixture – 0.2 wt% per aqueous phase - according to the approach described elsewhere.[5,6] This allowed to increase the conversion of monomers up to 91 wt% in S4 synthesis.

After HCl/NaOH treatment of S3 latex by the method described above, the particles had a positive ζ-potential only at pH < 7.0, which indicated their amphiphilic surface layer (Figure 3). Conductometric titration of weakly acidic and basic surface groups before and after HCl/NaOH treatment showed an increase in their total surface concentration (Table 1), which supported hydrophilization of the particle surface layer.

The ratio of surface amino and carboxylic groups can be tuned, varying the copolymerization conditions. Thus, near the CTAB critical concentration of micellization (CCM = 0.9 mmol/L at 25 °C) nanoparticles retained their positive surface charge from pH 3 to 10 as well as after the HCl/NaOH treatment (Figure 4a). This feature could be explained by the competition of CTAB cations and terminal cationic imidazoline groups of polymer chains for the place in the particle surface layers. At

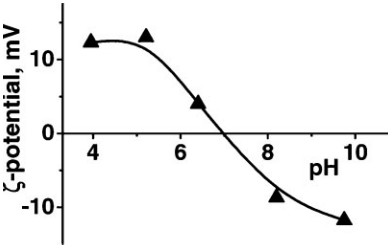


Figure 3.
ζ-potential of S3 particles after HCl/NaOH treatment (10^{-3} mol/L NaCl).

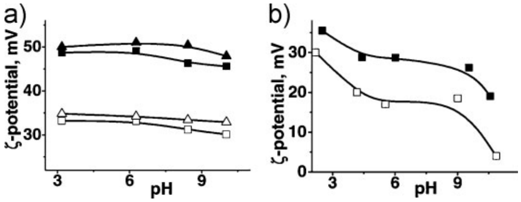


Figure 4.
ζ-potential of N1 (a) and N2 (b) latex nanoparticles; □, △ = 10^{-2} mol/L NaCl; ■, ▲ - 10^{-3} mol/L NaCl; □, ■ - without and △, ▲ - after HCl/NaOH treatment.

the CTAB concentration remarkably exceeding CCM, the surface density of AIP residues decreases, leading to a decrease in the concentration of surface carboxylic groups formed by hydrolysis of AIP residues. A similar behavior was observed earlier for polystyrene nanoparticles obtained by emulsion polymerization of styrene in the presence of sodium dodecyl sulfate and 4,4′-azobis(4-cyanoisovaleric acid). It was found that the anionic emulsifier ousted the carboxyl-containing azo initiator residues from the particle surface layer.[19]

Finally, the MMA - NVF copolymerization was successfully carried out in the presence of both CTAB and dextran to give monodisperse latex nanoparticles with not only hydrophilic and amphiphilic, but also biocompatible surface (N2 sample). Their ζ-potential dependences on pH and NaCl concentration (Figure 4b) resemble those of submicron particles of the same nature (S2 sample, Figure 2b), but the ζ-potential values were more positive in the weakly alkaline range.

At room temperature in dialyzed latexes of nanoparticles, the formation of periodic colloid structures was observed at polymer concentrations about 1–2 wt%. This manifested itself as iridescence of the whole dispersion volume or separate ordered structures with the cross-section square $\sim$5 mm^2. The dispersions were quite stable, although the ordered iridescent structures gradually diminished (without destruction) at the rate $\sim$1.5 cm per day. The formation of the iridescent structures is also reversible: they can be destroyed by shaking, but reassemble in 2 min after the interruption of mechanical disturbance. It is obvious that these large structures are multilayered lattices, which caused iridescence due to light diffraction. The reflection peaks in the transmission spectra of the nanoparticle dispersions were observable for about 30-60 min (Figure 5) as the ordered structures formed in the first minutes after the sample injection into a 0.1 mm thick cuvette. They were then oriented in parallel and finally precipitated on the walls of the cuvette.

For the systems under investigation, one should take into account the possibility of stretching of hydrophilic polymer chains from the particle surface into the bulk of the aqueous phase with the formation of interparticle entanglements and/or H-bonds. As these chains possess both amino and carboxylic groups, the interaction between amphiphilic particles could even of longer range than it was found by Cardoso et al.[20,21] It may be suggested that such cooperative interactions are able to provide a higher stability of the ordered structures of the obtained particles, making them perspective materials for creating various nanostructured matrices for nanotechnological applications.

Therefore, S3 and S4 latexes with submicron-sized monodisperse particles were tested as building blocks of hierarchic materials. Their self-assembling on glass slides into 3D-ordered arrays (Figure 6a) which showed photonic crystalline features was observed. In particular, these arrays displayed a predominant peak correspond-

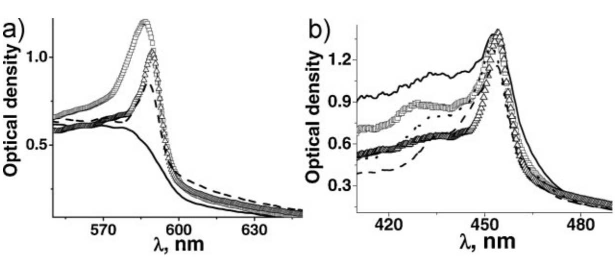


Figure 5.

Transmission spectra of N1 (a) and N2 (b) dialyzed latexes. Time of ageing: solid line - 0, ■ - 5, △ - 20, dash line - 30, dot line - 60 min.

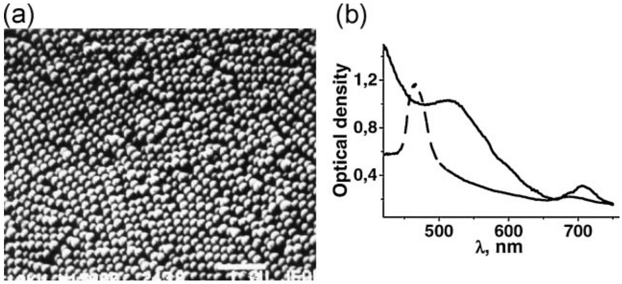


Figure 6.
SEM image of the film formed by S4 latex particles (a) and transmission spectra of solid thin films obtained by self-assembling of S3 (solid line) and S4 (dashed line) latex particles on glass slide (b).

ing to photonic band gap and originating from the Bragg diffraction of electromagnetic waves on 3D-ordered structures (Figure 6b).

Conclusion

One-step batch emulsion copolymerization of MMA or styrene with NVF initiated by 2,2′-azobis[2-(4,5-dihydroimidazol-2-yl) propane] dihydrochloride in the presence of dextran and/or low concentration of cetyltrimethylammonium bromide as well as without any stabilizers was shown to be a proper way to obtain monodisperse particles having a positive surface charge and hydrophilic particle surface. As a result, a narrow particle size distribution was obtained for particle diameters from 75 nm to submicron range. After hydrolysis of the obtained particles, their surface layers contained amino groups originating from NVF units along with carboxylic groups from hydrolyzed azo initiator residues. Their ratio can be varied by synthesis conditions. Self-assembling of the obtained amphiphilic latexes is a very facile process which occurs already in their dilute dispersions. An obvious advantage of these latex particles is that one can easily control their self-assembling by changing hydrophilicity of the surface layer and varying the surface concentration and ionization degree of ionogenic (carboxylic and amino) groups.

Acknowledgements: The work was supported by the Russian Federal Agency for Science and Innovations (contract 02.513.11.3166) and by the program U.M.N.I.K. (project 07/2-I-03).

[1] *Colloidal Biomolecules, Biomaterials, and Biomedical applications*, A. M. Elaissari, Ed., Dekker, New York **2004**, 488 p.
[2] *Colloids and Colloid Assemblies*, F. Caruso, Ed., Wiley-VCH, Weinheim **2004**, *621* p.
[3] T. Ruhl, P. Spahn, G. P. Hellmann, *Polymer*, **2003**, *44*, 7625.
[4] A. Yu. Menshikova, B. M. Shabsels, T. G. Evseeva, N. N. Shevchenko, A. Yu. Bilibin, *Russian J. Appl. Chem.* **2005**, *78*, 159.
[5] A. Yu. Menshikova, B. M. Shabsels, N. N. Shevchenko, A. G. Bazhenova, A. B. Pevtsov, A. B. Selkin, A. Yu. Bilibin, *Colloids Surf. A* **2007**, *298*, 27.
[6] B. Verrier-Charleux, C. Graillar, Y. Chevalier, C. Pichot, A. Revillon, *Colloid Polym. Sci.* **1991**, *269*, 398.
[7] T. Delair, V. Marguet, C. Pichot, B. Mandrand, *Colloid Polym. Sci.* **1994**, *272*, 962.
[8] M. Okubo, Y. Iwasaki, Y. Yamamoto, *Colloid Polym. Sci.* **1992**, *270*, 733.
[9] V. L. Covolan, F. Galembeck, C. A. Paula Leite, G. Ruggeri, E. Chiellini, *J. Colloid Interface Sci.* **2004**, *273*, 121.
[10] I. Miraballes-Martinez, A. Martin-Molina, F. Galisteo-González, J. Forcada, *J. Polym. Sci. A* **2001**, *39*, 2929.
[11] L. Gu, S. Zhu, A. N. Hrymak, *J. Appl. Polym. Sci.* **2002**, *86*, 3412.
[12] J. Xu, A. B. Timmons, R. Pelton, *Colloid Polym. Sci.* **2004**, *282*, 256.
[13] B. J. W. Goodwin, R. H. Ottewill, R. Pelton, G. Vianello, D. E. Yates, *Br. Polym. J.* **1978**, *10*, 173.
[14] B. J. W. Goodwin, R. H. Ottewill, R. Pelton, *Colloid Polym. Sci.* **1979**, *257*, 61.
[15] A. V. Yakimansky, A. Yu. Menshikova, T. G. Evseeva, N. N. Shevchenko, A. Yu. Bilibin, *Nanotechnol. Russia* **2006**, *1*, 171.
[16] A. Yu. Menshikova, T. G. Evseeva, N. A. Chekina, Y. O. Skurkis, S. S. Ivanchev, *Prog. Colloid Polym. Sci.* **2003**, *124*, 68.

[17] A. Yu. Menshikova, T. G. Evseeva, N. A. Chekina, S. S. Ivanchev, *Russian J. Appl. Chem.* **2001**, *74*, 489.

[18] A. V. Selkin, A. Yu. Bilibin, A. Yu. Menshikova, Yu. A. Pashkov, N. N. Shevchenko, A. G. Bazhenova, *Bull. Russian Acad. Sci. Phys.* **2005**, *69*, 1111–1112.

[19] A. Yu. Menshikova, T. G. Evseeva, B. M. Shabsels, A. Yu. Menshikova, T. G. Evseeva, B. M. Shabsels, I. V. Balanina, A. K. Sirotkin, S. S. Ivanchev, *Russian J. Appl. Chem.* **2005**, *78*, 1008.

[20] A. H. Cardoso, C. A. P. Leite, M. E. D. Zaniquelli, F. Galembeck, *Colloids Surf. A* **1998**, *144*, 207.

[21] A. H. Cardoso, A. Carlos, F. Galembeck, *Langmuir*, **1999**, *15*, 4447.