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Photo-Cross-Linked Diblock Copolymer Micelles: Quantitative Study of Photochemical Efficiency, Micelles Morphologies and their Thermal Behavior

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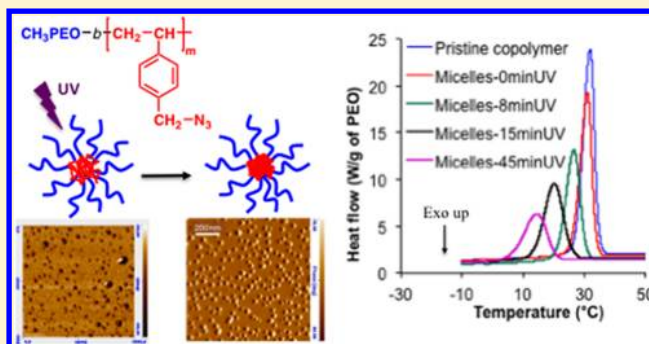
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S Supporting Information

ABSTRACT: Amphiphilic block copolymers based on azido-functional polystyrene (PVBA) and hydrophilic poly(ethylene oxide) (PEO) have been synthesized by nitroxide mediated polymerization and used to prepare aqueous spherical micelles. These micelles consisting of a PEO corona and a PVBA core were stabilized by cores cross-linking under UV irradiation without addition of photoreagent. The efficiency of the core cross-linking was estimated in term of the conversion of azide groups to dibenzylidene junctions with liberation of gaseous nitrogen molecules. The micellar structures were studied by dynamic light scattering (DLS) and atomic force microscopy (AFM). Differential scanning calorimetry (DSC) was used to characterize the thermal behavior of freeze-dried micelles. Attachment of PEO block to a rigid core modified considerably the thermal behavior of PEO. We observed a decrease of about 15 °C for the crystallization temperature and 50% for the degree of crystallinity of cross-linked micelles submitted to 45 min UV irradiation compared to pristine copolymer.



INTRODUCTION

Whenever amphiphilic block copolymers comprising hydrophilic and hydrophobic blocks are dissolved in an aqueous solution, they can self-assemble into supramolecular assemblies, characterized by core-shell structures, called polymeric micelles. Polymeric micelles have found wide-range applications including nanodelivery systems (i.e., for depot drug release and targeted drug delivery),^{1,2} sensor,³ template for mesoporous silica synthesis,⁴ filtration membrane,⁵ and catalysis.⁶

Although polymeric micelles are quite stable compared to their homologue low molecular weight surfactants, they also are always in thermodynamic equilibrium with their constituent unimers and are sensitive to the environment such as at low concentrations, at high temperature or under certain changes in solvent conditions. This sensitivity can lead to a lack of micelles stability and their disintegrations. In the past decades, the strategies of covalent stabilization via cross-linking of the micellar core or shell were explored extensively to overcome the instability problem.⁷ The mostly proposed applications of cross-linked polymeric micelles are drug and gene delivery. The principal reasons to cross-link micelles for drug delivery applications are to prevent premature release of the active compound, to introduce controlled and/or stimuli responsive

release properties, or to stabilize micelles *in vivo*. There are several papers reviewing in details drug delivery applications.^{8–10}

The field of shell cross-linked micelles was initiated by the group of Wooley.¹¹ Shell cross-linked micelles include those based on AB diblock and ABC triblock copolymers. However, cross-linking of hydrophilic shells may affect the surface mobility and a highly diluted solution is often required to avoid the formation of intermicellar cross-linking.¹² For the core cross-linking, there are several potential locations, including at the core chain end, within the core domain or at the core-shell interface. The usual chemistries employed for core cross-linking and stabilization of the micellar particles are as follows: the incorporation of polymerizable or photo/UV-cross-linkable groups,^{13–21} the introduction of cross-linking reagents,^{22–25} and the introduction of external stimuli.^{26,27} Among these different approaches, the photo-cross-linking chemistry presents significant advantages since no byproducts are formed during the photo-cross-linking procedure. Fur-

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thermore, the majority of micellar characteristic features, such as micelles size and shape are maintained during the photo-cross-linking process and the cross-linking density can be easily controlled by tuning the light energy or exposure time. All of those advantages make the photo-cross-linking chemistry a good choice to tune the stability of micelles. To achieve such photo-assisted cross-linking, the most commonly used reactions are photocycloaddition and photopolymerization. For instance, reversible dimerization by intermolecular photocycloaddition of core-forming blocks containing thymine,¹⁸ coumarin,^{16,19} or cinnamate side groups^{28–32} was essentially used.

In the case of photopolymerization, any core-forming polymer blocks that contain double bonds such as aliphatic double bonds, vinylic or acrylic moieties are prone to micelles stabilization by UV cross-linking. These double bonds are present either within the polymer main chain, as terminal chain ends, or as pendant side groups. In most of these cases, a photoinitiator was added during the micellization process and then UV light was applied to initiate the cross-linking of micelles core. This concept was first introduced by Prochaska et al. more than 30 years ago in the photo-initiated UV polymerization of polybutadiene.³³ Recently, one or several cross-linkable end groups such as methacryloyl can also be attached to the block copolymers and undergone photopolymerization in the presence of a UV initiator. For example, Nicol et al.¹⁴ synthesized an asymmetric poly(ethylene oxide)-*b*-poly(ethyl acrylate) block copolymers bearing polymerizable (meth)acrylate groups at the end of the hydrophobic block. Photocross-linking process was induced by using a photo-initiator (2,2-dimethoxy-2-phenylacetophenone) and a cross-linker (butandiol dimethacrylate). They demonstrated that a complete reaction of double bonds has been achieved within a few seconds without modification of the structural properties of the micelles. Other examples of block copolymers containing double bonds were also studied but the novelty of the works cited below is that photo-cross-linking was simply produced by exposing micellar solution to UV irradiation without addition of UV initiators. Indeed, Gohy et al.¹⁵ reported the core cross-linked aqueous spherical micelles from amphiphilic poly(2-ethyl-2-oxazoline-*block*-2-“soy alkyl”-2-oxazoline) diblock copolymers. The hydrophobic block contained on average 1.4 double bonds in each alkyl chain. Whereas, Chang et al.²⁰ reported the preparation of carbon nanospheres by pyrolysis of core cross-linked diblock copolymer micelles. The diblock copolymer in this article was a poly(ethylene oxide)-*b*-poly(4-ethynylstyrene) with a hydrophobic block having curable acetylenic groups. The authors presumed that the photo-reaction proceeded during the UV irradiation was an oxidative coupling between the acetylenic groups.

The stabilization of polymeric micelles via cross-linking reactions is an important aspect of the application of these materials.⁹ However, cross-linking chemistries are inherently inefficient and hard to quantify experimentally. Thus, the development of new cross-linking strategies is still an important objective in the application and development of stabilized polymer micelles.

Nowadays, amphiphilic copolymers bearing pendant azide groups in the hydrophobic segment were only used for core cross-linking of micelles by click chemistry.^{24,34,35} Nevertheless, it was reported that polymers containing azide side groups could be bulk cross-linked by thermal treatment or by UV light irradiation.^{36–38} Click reaction used for core cross-linking of micelles need the addition of reagent bearing alkyne group,

while the cross-linking of azide side groups by thermal treatment or by UV light irradiation do not need addition of any reagent. In the present work, we report on the synthesis of cross-linkable diblock copolymers poly(ethylene oxide)-*b*-poly(vinylbenzyl azide) (PEO-*b*-PVBA) using nitroxide mediated polymerization (NMP). Then these materials have been used to generate aqueous spherical micelles and performed their subsequent covalent core cross-linking by photoinduced cleavage of azide groups. Efficiency of the photochemical reaction is easily quantified as a function of UV irradiation time thanks to elemental analysis of nitrogen. Thermal properties of freeze-dried micelles are deeply characterized by differential scanning calorimetry (DSC) showing a strong influence of cross-linking degree on the crystallization of PEO block.

■ EXPERIMENTAL SECTION

Materials. 4-Vinylbenzyl chloride (90%, VBC), NaN₃ (>99.5%) and poly(ethylene glycol) methyl ether (CH₃-PEO) with a molar mass of 5000 and 20000 g·mol⁻¹ were all obtained from Aldrich and used as received. BlocBuilderMA (>99%), an alkoxyamine based on the nitroxide SG₁ (*N*-*tert*-butyl-*N*-[1-diethylphosphono(2,2-dimethyl-propyl)] nitroxide) and the 1-carboxy-1-methylethylalkyl moiety was kindly provided by Arkema (France). All solvents and other reagents were synthesis grade and used without further purification.

Poly(ethylene oxide)-*b*-poly(vinylbenzyl chloride) (PEO-*b*-PVBC) Copolymer Synthesis. The synthesis of diblock copolymer consisted in three steps. The first two steps aim to synthesize CH₃-PEO-macroinitiator and were performed following previous reported procedures.^{39,40} Briefly, the first step is the esterification of the CH₃-PEO-hydroxy with acryloyl chloride to form the corresponding PEO-acrylate. The second step corresponds to the 1,2 intermolecular radical addition of the BlocBuilderMA⁴¹ onto the acrylate group to yield the corresponding PEO-based macroalkoxyamines. The third step is the nitroxide-mediated polymerization of 4-vinylbenzyl chloride using PEO-based macroalkoxyamine as initiator. Typically, 4-vinylbenzyl chloride (2 g, 11.8 mmol) and PEO-based macroalkoxyamine (2 g, *M*_n = 5436 g·mol⁻¹, 0.36 mmol) were dissolved in 4g of ethylbenzene. The mixture was then loaded in a three-neck flask equipped with a reflux condenser and a magnetic stir bar, purged for 20 min at 40 °C to remove oxygen. Polymerization was performed at 110 °C for 150 min under argon atmosphere. The crude product was dissolved in dichloromethane and then poured into a large excess of cold diethyl ether/*n*-pentane mixture (50/50 v/v) to precipitate copolymer. The copolymer was then isolated by filtration and dried under vacuum at room temperature to a constant weight. Similar process was applied for the synthesis of the second diblock copolymer consisting of PEO of molar mass of 20000 g·mol⁻¹. The synthesized block copolymers were called **P1** and **P2** respectively. Their molar masses and compositions were determined by size exclusion chromatography (SEC) and proton nuclear magnetic resonance (NMR) spectroscopy.

Poly(ethylene oxide)-*b*-poly(vinylbenzyl azide) (PEO-*b*-PVBA) Copolymer Synthesis. In a typical experiment, the PEO-*b*-PVBC diblock copolymers (2g) was dissolved in 15 mL of *N,N*-dimethylformamide (DMF) in a round-bottom flask. NaN₃ (3 eq to chloride group) was added in one portion. The resulting solution was stirred at room temperature for 24 h. The reaction mixture was then diluted with dichloromethane. The product was isolated by precipitation in cold diethyl ether/*n*-pentane mixture (50/50 v/v) and dried under vacuum at room temperature. The resulting block copolymers containing azide groups were called **P1-N₃** and **P2-N₃**, respectively.

Micelles Preparation and UV Cross-Linking of the Cores. Micelles were prepared by dissolving PEO-*b*-PVBA copolymer in tetrahydrofuran (THF) at a concentration of 0.5% (w/v) for **P1-N₃** and 0.02% (w/v) for **P2-N₃**. Water was added dropwise to trigger micellization. THF was then eliminated by dialysis (Spectra Por Dialysis membranes with a MW cutoff of 1000 g/mol) against water

during 5 days. The concentration of the micelles in water was set between 0.4 and 0.015 wt % after the dialysis process. Fifteen mL of micellar solutions of block copolymers were cross-linked by applying UV-light (Sylvania F15Wx16 UV lamps, $\lambda_{\text{max}} = 350$ nm) at 23 °C under magnetic stirring for different time duration. The UV-lamps were kept at a distance of 10 cm from the sample surface. Cross-linked copolymer micelles were first studied in aqueous medium and then recovered by freeze-drying to study the system in organic medium and to determine the photochemical reaction yield.

Characterization Techniques. ^1H NMR spectra in CDCl_3 were recorded on a Bruker Advance 400 spectrometer. Chemical shift was given in ppm relative to tetramethylsilane.

Polymer molecular weights and dispersities were determined by size exclusion chromatography (SEC). The used system was an EcoSEC (Tosoh, Japan) equipped with a PL Resipore Precolumn (4.6×50 mm, Agilent) and two linear M columns (4.6×250 mm, Agilent) with a gel particle diameter of 3 μm . These columns were thermostated at 40 °C. Detection was made by an UV/visible detector operated at $\lambda = 254$ nm, a dual flow differential refractive index detector, both from Tosoh, and a viscometer ETA2010 from PSS. Measurements were performed in THF at a flow rate of 0.3 mL min^{-1} . Calibration was based on polystyrene standards from Polymer Laboratories (ranging from 370 $\text{g}\cdot\text{mol}^{-1}$ to 371 100 $\text{g}\cdot\text{mol}^{-1}$).

Dynamic light scattering (DLS) measurements were performed on a Malvern Nanosizer ZS-apparatus equipped with a He–Ne laser operating at a wavelength of 633 nm. The temperature was controlled at 25 °C. The method of the cumulants was generally used to analyze DLS results, while size distribution histograms were obtained by the CONTIN method. The polydispersity index (PDI) of the micelles was estimated from the ratio Γ_1/Γ_2 in which Γ_1 and Γ_2 represent the first and second cumulant, respectively.

Elemental analyses were made in order to determine the percent of nitrogen in micelles block copolymers before and after UV irradiations. Analyses were conducted by Spectropole analytical service (Marseille) on the Thermo Finnigan EA 1112 analyzer.

Samples for atomic force microscopy (AFM) were prepared by spin-coating of 0.1 mL of sample solution onto precleaned silicon plates (1×1 cm). The spin coater turned on at 2000 rpm for 60 s with a speed ramp of 50 $\text{rpm}\cdot\text{s}^{-2}$. AFM measurements were obtained at ambient conditions with a scanning probe microscope (JEOL JSPM-5410) equipped with commercially available Si-cantilevers (Budget Sensors, Tap 300 Al-G) with resonance frequency around 300 kHz and force constant of 20–75 $\text{N}\cdot\text{m}^{-1}$, according to the manufacturer. Height and phase images were recorded in soft tapping mode, with a scan speed of 1 Hz, to minimized interactions between the tip and the sample.

Differential scanning calorimetry (DSC) was carried out on a TA DSC Q20 using a heat/cool/heat cycle from -90 to $+130$ °C. After the cooling cycle, samples were kept at -90 °C for 30 min before being heated to 130 °C. The heating and cooling rates were 5 °C $\cdot\text{min}^{-1}$. Because the sample thermal history alters the obtained results, the first cycle was used to obtain samples with the same thermal history (similar crystallization conditions); afterward, the second and next cycles are reproducible.

RESULTS AND DISCUSSION

Synthesis of PEO-*b*-PVBA Diblock Copolymers. The success of the NMP of various vinylic monomers using alkoxyamine BlocBuilderMA or its derivatives as initiators was widely reported.⁴² This technique was therefore used for the preparation of PEO-*b*-PVBC block copolymers using the corresponding PEO-based macroalkoxyamine as initiator. The ^1H NMR analyses of the PEO-*b*-PVBC copolymers were performed to determine their composition. Typically, knowing the molar mass (M_n) of the starting PEO, the experimental M_n of the poly(4-vinylbenzyl chloride) block was calculated from ^1H NMR spectra of the resulting copolymer, based on the integration of $-\text{CH}_2-\text{Cl}$ protons peak of PVBC block

normalized by this of ethylenic protons ($\delta = 3.4$ ppm) of PEO. The obtained data are given in Table 1. Then chloride

Table 1. Molar Mass Data of the Synthesized Amphiphilic Block Copolymers

sample	$M_{n,\text{PEO}}$ (g/mol)	$M_{n,\text{total}}^a$ (g/mol)	$M_{n,\text{total}}^b$ (g/mol)	dispersity (\bar{D}) ^b
P1	5000	8000	7400	1.07
P1- N_3	5000	8100	7500	1.07
P2	20 000	27 800	14 500	1.05
P2- N_3	20 000	28 100	18 300	1.14

^aValues determined by ^1H NMR spectroscopy. ^bSize exclusion chromatography.

groups of PVBC were substituted by azide groups via the reaction with NaN_3 . The ^1H NMR spectra (Figure 1) before and after substitution reaction clearly indicate the shift of methylene protons in α of the chloride group at 4.50 ppm (H8, Figure 1A) to 4.20 (H8', Figure 1B), demonstrating that complete substitution occurred. The same conclusions could be drawn from the reaction between P2 and NaN_3 . Block copolymers were also characterized by SEC. The M_n and dispersity (\bar{D}) values derived from a calibration curve based on PS standards are summarized in Table 1. With such calibration, only \bar{D} can be reasonably extracted from the chromatograms whereas the values of the total M_n cannot be trusted. Figure 2 shows the SEC traces of the PEO-based alkoxyamines and the corresponding P1- N_3 and P2- N_3 diblock copolymers. The SEC chromatogram of P1- N_3 was monomodal without any tailing (at high elution volume) potentially originated from residual PEO macroinitiator or any shoulder (at low elution volume) that could be caused by transfer and/or termination reactions. On the other hand, the SEC chromatogram of P2- N_3 was less symmetrical, indicating partial termination or chain transfer during the polymerization. This suggests that P1- N_3 copolymer having a better well-defined architecture than that of P2- N_3 copolymer.

Micellization of Block Copolymers and Cross-Linking of the Micelles Cores. Since they contain a major hydrophilic PEO block, the P1- N_3 and P2- N_3 samples are good candidates for the formation of aqueous micelles. Samples were first dissolved in THF, which is qualified as a good solvent for poly(vinylbenzyl azide) block and a medium solvent for PEO block. For this reason, P2- N_3 copolymer containing a longer PEO block could only be dissolved at a maximum concentration of only 0.02% (w/v) in this solvent. Whatever the copolymers, DLS confirmed that the copolymer chains were molecularly dissolved in THF. Water was then added dropwise to trigger micellization process. This procedure has the advantage to ensure an initial complete solubilization of the block copolymers as unimers and to erase any memory effect of the bulk structure.⁶ THF was then gradually removed by dialysis against water. The micelles in pure water were characterized by DLS and AFM. One single population of micelles with an average diameter of 23 nm for P1- N_3 and 71 nm for P2- N_3 was found by DLS (Table 2 and Figure 3). As expected, longer copolymer chains (P2- N_3) product larger micelles. Micelles morphology was also visualized by AFM, and the obtained images (Figure 4) show spherical and stretched micelles for the two studied samples. Different populations were observed and their sizes D_x in x -direction (length) are generally larger than that measured by DLS, however, the sizes

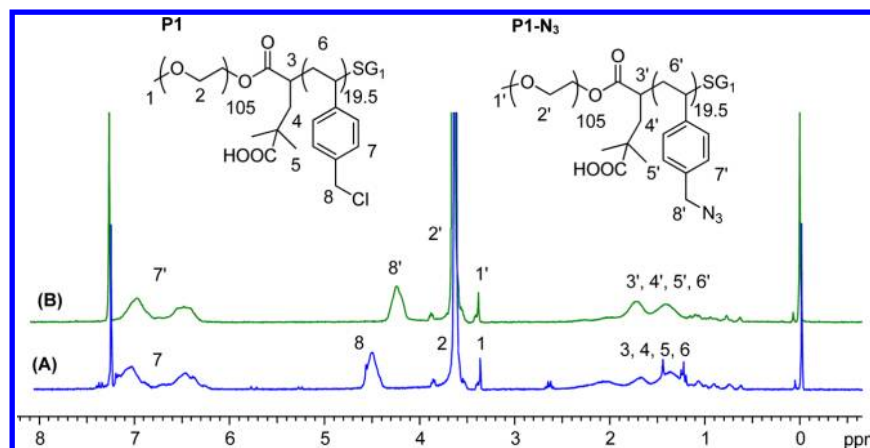


Figure 1. ^1H NMR spectra of (A) poly(ethylene oxide)-*b*-poly(vinylbenzyl chloride) **P1** and (B) poly(ethylene oxide)-*b*-poly(vinylbenzyl azide) **P1-N₃** in CDCl_3 .

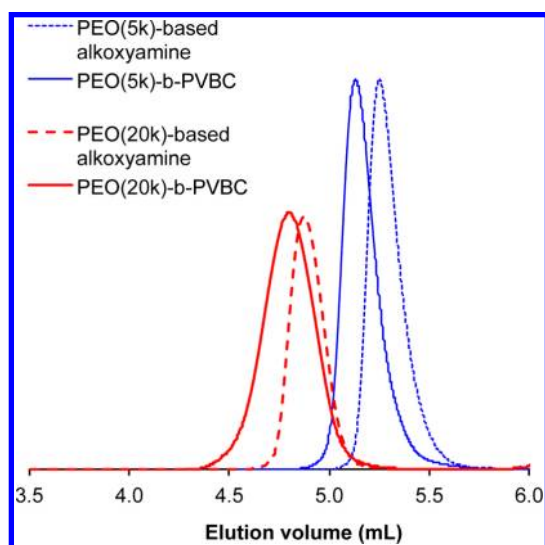


Figure 2. SEC traces in tetrahydrofuran of PEO-based alkoxyamines (dashed lines) and PEO-*b*-PVBC block copolymers (full lines).

Table 2. Characteristics Features of the PEO-*b*-PVBA Micelles before and after UV Irradiation As Measured by DLS and AFM

samples	DLS		AFM	
	Size (nm) ^a	PDI ^b	D_x (nm) ^c	D_z (nm) ^c
P1-N₃ micelles	23	0.053	28	6.2 ± 0.8
P1-N₃ micelles irradiated for 45 min	16 220	0.56	20	4.1 ± 0.5
P2-N₃ micelles	71	0.059	20	15.6 ± 1.3
P2-N₃ micelles irradiated for 45 min	69	0.14	20	20.4 ± 0.7

^aDiameter of micelles as measured by DLS. ^bPDI is the polydispersity index of the micelles as measured by DLS. ^c D_x and D_z are respectively the diameters of the micelles in the x -direction (length) and z -direction (height) as measured by AFM.

D_z in z -direction (height) are smaller (Table 2). This difference of morphology and sized observed in AFM measurements

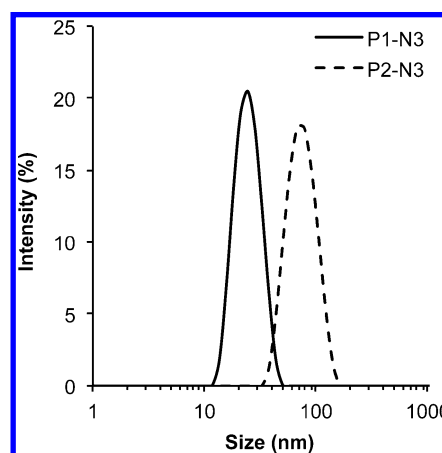


Figure 3. Size distribution by DLS of non-cross-linked micelles of two different block copolymers in water.

compared to that observed in DLS analysis can be explained by micelles fusion and flattened by the centrifugation forces during micelles application on silicon wafer by spin-coating. Indeed, it is well-known that polymeric micelles are a dynamic system where small perturbations (pH, temperature, solvents, ...) could change micelles shape and size.^{15,43}

In order to stabilize the micellar structures by cross-linking the cores, azide groups were introduced onto the hydrophobic block, which constitutes the micelles core. As reported, cross-linking of polymers containing azide side groups can be obtained thermally or by UV light irradiation with photocleavage of azide.^{36–38} Because of the instability of micelles with temperature change, we chose, in this work, UV assisted cross-linking of the micelles cores. The cross-linking step was performed by exposing the samples to UV light for a given time under stirring. During irradiation, samples were refrigerated at 23 °C. According to the literature,⁴⁴ the possible cross-linking mechanism for polymers containing azide as a pendent group is a result of the chemical reaction as illustrated in Scheme 1. The azene groups could be formed both by intermolecular and intramolecular reactions. However, because of the short PVB-N₃ block, particularly in **P1-N₃** copolymer, the intermolecular reaction is probably favored. Under UV irradiation, the azide groups in the polymer chains start to break, cleave off gaseous nitrogen and form an azo bond. As a result of this reaction, the chains of the cross-linked copolymer are joined by 1,2-

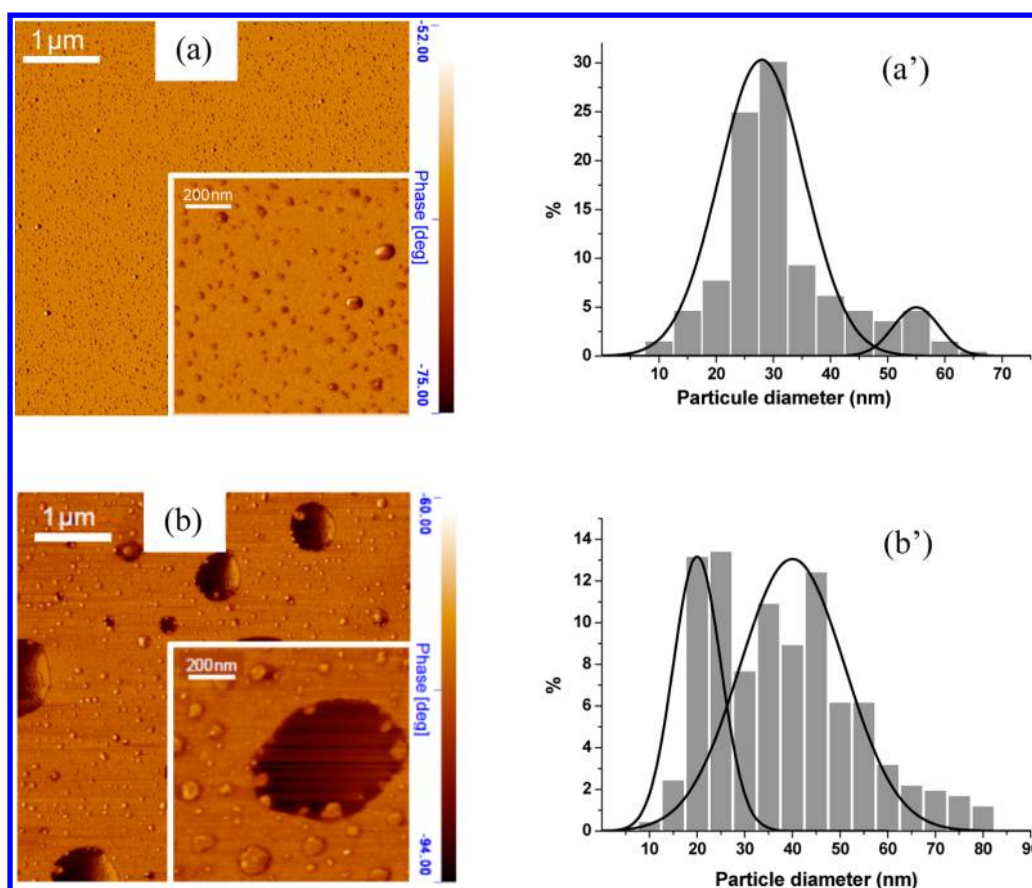


Figure 4. AFM phase images of non cross-linked micelles of (a) P1-N_3 and (b) P2-N_3 diblock copolymer. The histograms (a') and (b') correspond respectively to P1-N_3 and P2-N_3 diameter distributions. The curves (full line) are the fit of the first Gaussian function.

dibenzylidiazene groups (Scheme 1). Since each reaction between two azide groups liberates two gaseous nitrogen molecules, knowing the amount of nitrogen remaining in the copolymer micelles after UV irradiation, we can calculate the conversion of azide groups into azene groups. Irradiated samples at different times were freeze-dried and the amount of nitrogen was determined by elemental analysis. The conversion as a function of the exposure time is shown in Figure 5a for the cross-linking of P1-N_3 micelles.

As expected, conversion of azide to azene groups increased with irradiation time and reached a maximum value of 80% after 150 min of UV irradiation. For an irradiation duration between 2 and 8 min, DSL measurements did not show any formation of aggregates during the UV irradiation process and micelles size remained unchanged (Figure 1S, Supporting Information). However, big aggregates began to form when irradiation time was longer than 8 min (Figure 5b). Between 15 and 45 min of irradiation, P1-N_3 micelles exhibited a small increase of big aggregates in the range of size between 100–600 nm, which represent only 0.1% in volume of micellar solution (Figure 5c). The number and size of these large aggregates did not vary for longer irradiation time (i.e., 150 min). Formation of big aggregates was already observed by Gohy et al.¹⁵ during the cross-linking of micelles by UV light irradiation. Indeed, the large objects were observed in AFM images when the micellar solution of poly(2-ethyl-2-oxazoline-*block*-2-“soy alkyl”-2-oxazoline) was exposed to high doses of UV light. They suggested that the clustered micelles were formed by intermicellar cross-linking via the chain exchange dynamics of initial uncross-linked

micelles. According to Tenhu and co-workers, chain exchange between micelles could occur via two ways: (i) insertion and expulsion of single chains and (ii) merging and splitting of micelles.⁴⁵ In our system, the latter is probably the operative mechanism.

It was also observed that for UV irradiation duration longer than 8 min is accompanied by a slight decrease of the micelles size (−3 to −5 nm of diameter), and then stabilized beyond 45 min of UV irradiation while the conversion of azide to azene groups continues to increase (Figure 5). This variation of micelles size is mostly due to the shrinkage of the micelles core. Indeed, the photochemical reaction of the cross-linking process in this study generated a lost of one gaseous nitrogen molecule per styrene monomer unit. Less than 8 min of irradiation (<10% of conversion), the lost of gaseous nitrogen was low and did not affect the core size. When the exposure time was sufficiently long (15–45 min with 20–40% of conversion), the lost of gaseous nitrogen became significant as well as the cross-linked network within the micelles core that was now tightly packed. This feature contributes to shrink the micelles core leading to a decrease of the micelles size. Beyond 45 min of irradiation, since the micelles core is now sufficiently cross-linked and stabilized, a supplementary lost of gaseous nitrogen did not affect the micelles size and shape. In order to check the stabilization of irradiated micelles during the storage in aqueous medium, DLS measurements of micelles irradiated for 45 min were performed at different storage times. The obtained results show that the micelles sized slightly increased (+2 nm of diameter) after one day of storage and then reached

Scheme 1. Cross-Linking of PEO-*b*-PVBA Block Copolymer by UV Irradiation at a Wavelength of 350 nm with the Formation of Azene Groups and Production of Gaseous Nitrogen According to (a) Intermolecular Reaction and (b) Intramolecular Reaction

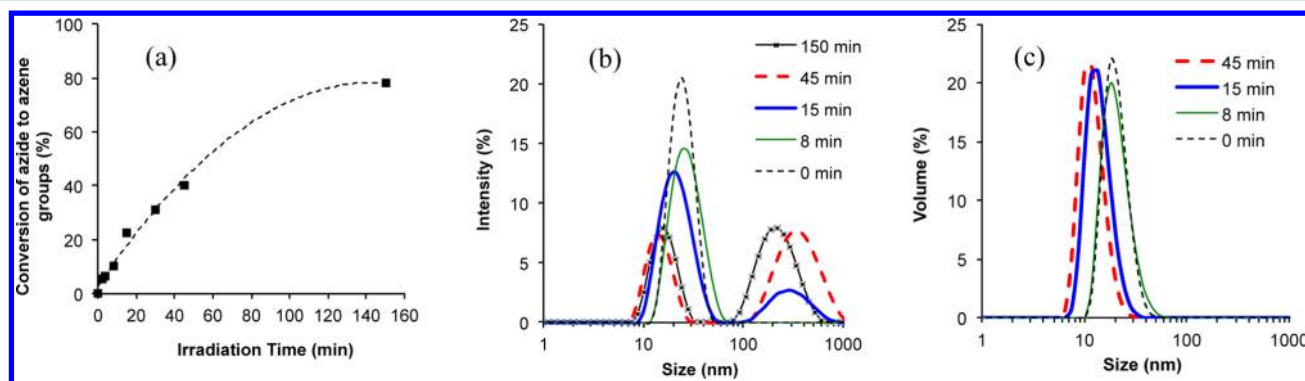
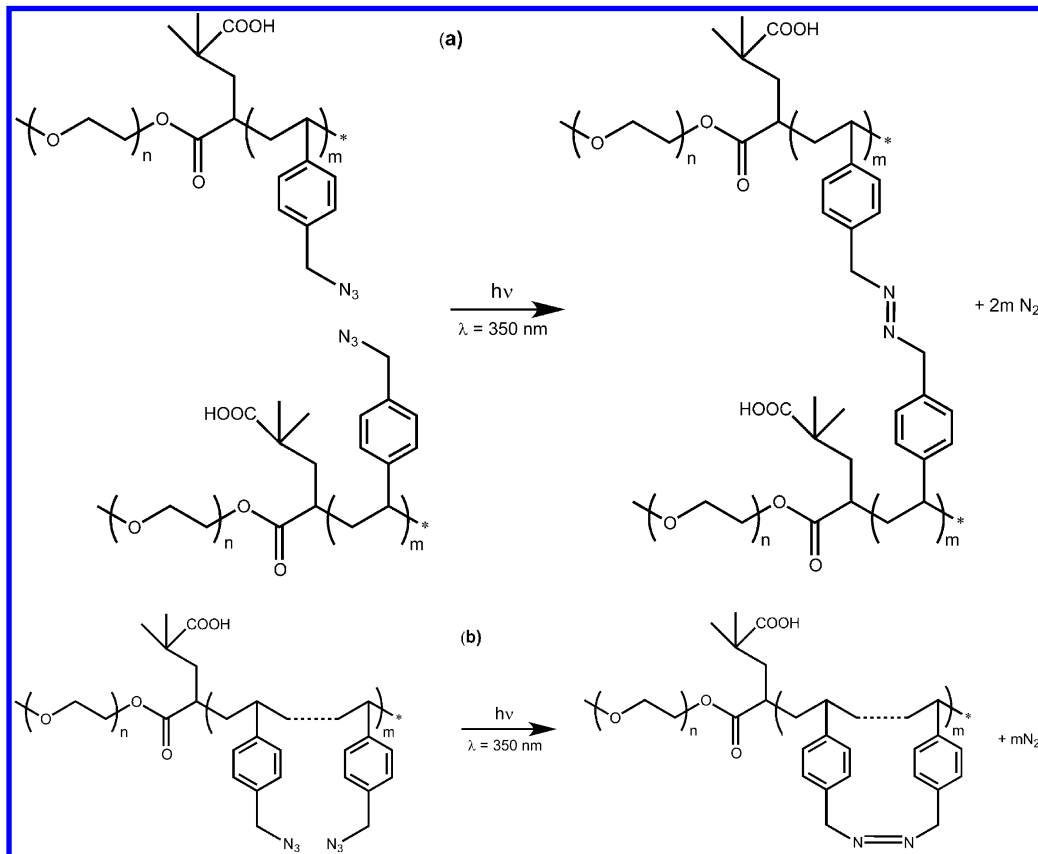


Figure 5. (a) Conversion of azide groups to azene groups as a function of irradiation time of **P1-N₃** micelles. Particles size distribution of cross-linked **P1-N₃** micelles at different irradiation durations by DLS in water; data shown in (b) intensity and (c) volume.

equilibrium and remained unchanged for the remaining of time (Figure 2S). This small variation in micelles size is probably due to the diffusion of some water molecules inside the micelles core that slightly swells it.

Contrary to **P1-N₃** micellar solution, that of **P2-N₃** at a concentration of 0.015 wt % irradiated for 45 min did not show any large aggregates. We also noted a very slight decrease of micelles size after irradiation but the decrease was less significant than that observed for **P1-N₃** micelles (Figure 3S). This difference observed between two copolymers is probably due to the copolymer blocks length and the micellar solution concentration. Indeed, **P1-N₃** is shorter than **P2-N₃**, the core of **P1-N₃** might not be well shielded by the PEO

chains and the block segregation might not be so sharp. In addition, the core chains could readily pull out of existing micelles and possessed high mobility and in consequence, these micelles could fuse during core cross-linking. On the other hand, micellar solution was more concentrated than that of **P2-N₃** micellar solution, thereof, in a same volume, the number of **P1-N₃** micelles should be higher than that of **P2-N₃** micelles even if the size of the latter is three times bigger. As a result of this, **P1-N₃** micelles are close enough to chain exchange and produce large aggregates. This hypothesis was evidenced by preparing different **P1-N₃** micellar solutions at a concentration of 0.02, 0.05, and 0.2 wt % and then irradiated them by UV light for 45 min. Indeed, we observed a strong

reduction of big aggregates with the decrease of micellar solution concentration (Table 3). These big objects have totally disappeared in the aqueous P1-N_3 micellar solution at a concentration of 0.02 wt % and irradiated for 45 min (Figure 5S).

Table 3. Variation of the Percentage of Big Aggregates as a Function of the Micellar Solution Concentration in P1-N_3 Micelles Irradiated for 45 min As Measured by DLS

micellar solution concentration (wt %)	micelles size (nm)		
	before UV irradiation (PDI)	45 min UV irradiation	
		different aggregates (PDI)	% in intensity
0.5	23 (0.053)	16 (0.56)	58
		220	42
0.2	25 (0.058)	18 (0.32)	84
		180	16
0.05	26 (0.061)	25 (0.24)	90
		89	10
0.02	27 (0.062)	25 (0.014)	100

AFM was also used to visualize the micelles morphology after photo-cross-linking process. AFM images of cross-linked micelles of P1-N_3 and P2-N_3 copolymers irradiated during 45 min are shown in Figure 6. Spherical structures of almost

micelles maintained intact after UV cross-linking contrary to uncross-linked micelles which merged under effect of centrifugation forces during sample preparation for AFM analysis by spin-coating (Figure 3). This indicates that the photo-cross-linking of micelles based on PEO block copolymers was successful via the UV irradiation of azide side groups grafted on hydrophobic block. Dimensional characteristics of cross-linked micelles after 45 min UV irradiation are summarized in Table 2. It is not surprising that one observed differences in absolute measurements between DLS and AFM because DLS provides a measure of hydrodynamic dimensions while AFM is a direct measure of dried objects.

To validate the efficiency of the cross-linking process and confirm the stability of the cross-linked micelles, the P1-N_3 micelles irradiated for 8 and 15 min were first freeze-dried and then redispersed respectively in DMF and in dichloromethane (good solvents of both blocks) at a concentration of 0.05% (w/v). DLS measurements indicated the presence of micelles in these solvents, confirming that the micelles structure initially formed in water was stabilized by the photo-cross-linking process (Figure 4S). However, the hydrodynamic radius of the micelles was smaller in DMF than in water indicating a poorer solvation of the PEO chains in this solvent.

Thermal Behavior of PEO Shells in Diblock Copolymer Micelles with Cross-Linked Cores. To evaluate the impact of the degree of internal cross-linking on the thermal properties

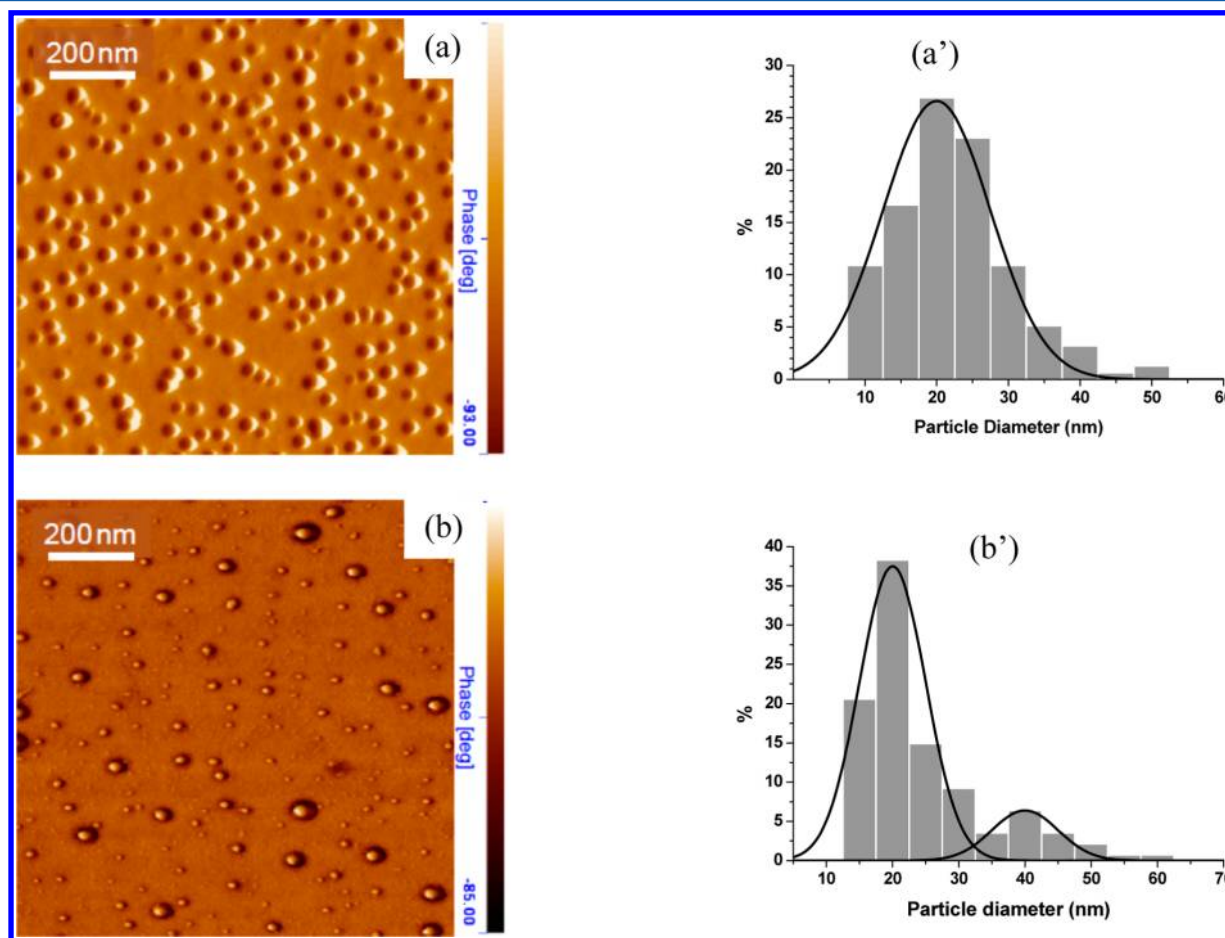


Figure 6. AFM phase images of cross-linked micelles irradiated for 45 min of (a) P1-N_3 and (b) P2-N_3 diblock copolymer. The histograms a' and b' correspond respectively to P1-N_3 and P2-N_3 cross-linked micelles diameter distributions. The curves (full line) are the fit of the first Gaussian function.

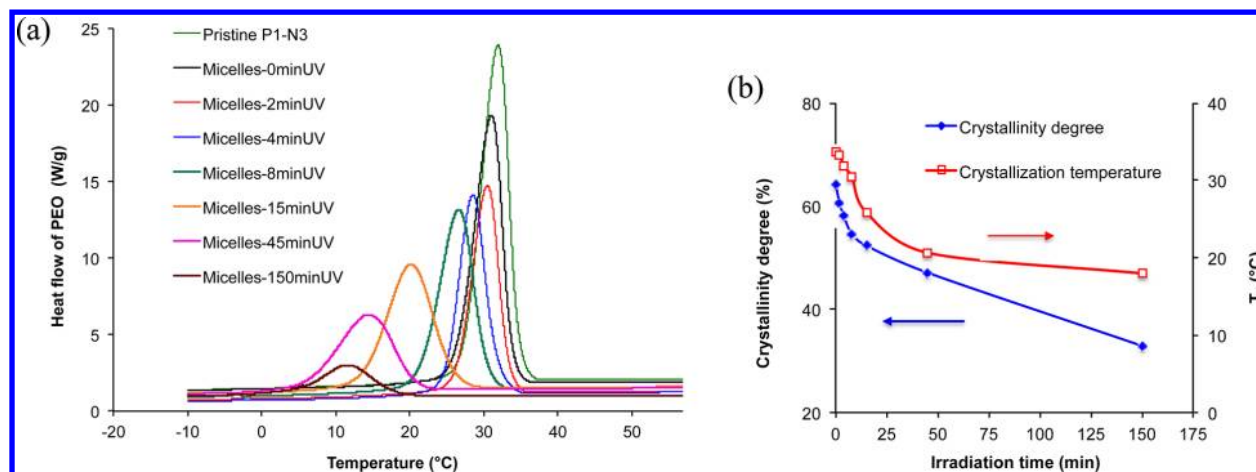


Figure 7. (a) DSC crystallization thermograms of the pristine **P1-N₃** copolymer and its micelles before and after UV irradiation obtained with a scanning rate of 5 °C/min under nitrogen. (b) Evolution of the degree of crystallinity and crystallization temperature of PEO as a function of UV irradiation time.

(melting and crystallization) of PEO, DSC experiments were performed for **P1-N₃** pristine block copolymer, **P1-N₃** micelles before and after UV irradiation. All samples were submitted to the same thermal treatment during DSC analysis (see Experimental Section). As for sample preparation history, **P1-N₃** micelles were obtained by freeze-drying (or lyophilization) of aqueous micelles solution and **P1-N₃** pristine copolymer was used as obtained after synthesis. The melting temperature, T_m , and the crystallization temperature, T_c , of PEO micelles shell were determined as the intersection of the baseline with the tangent at the inflection point of the melting and crystallization peaks, respectively. The thermograms obtained for the crystallization of PEO in the nonirradiated and irradiated **P1-N₃** micelles as a function of UV irradiation time are given in Figure 7a. The values of T_m , T_c , and the measured melting and crystallization enthalpies as well as the degree of crystallinity, X_c , are summarized in Table 1S. The degree of crystallinity was calculated by dividing the measured melting enthalpy by that of 100% crystalline PEO, $\Delta H_m^\circ = 195$ J/g.⁴⁶ The evolution of the PEO crystallization peaks in Figure 7 clearly shows that both the crystallization temperature and the degree of crystallinity of PEO chains in cross-linked micelles are strongly depressed with UV irradiation time, but the melting temperature of PEO block is less affected until 15 min exposure, and then decreased when the cross-linking degree reached significant value (i.e., 30% for 30 min UV irradiation). For sample irradiated during 150 min, where the degree of crystallinity was strongly reduced ($X_c = 32\%$), we observed a glass transition temperature (T_g) at -53.5 °C.

Decreasing of melting temperature was first observed by Wooley et al.⁴⁷ for poly(ϵ -caprolactone)-*b*-poly(acrylic acid) (PCL-*b*-PAA) core-shell micelles in which PCL constitutes the crystalline core domain and the PAA shell is cross-linked by 2,2'-(ethylenedioxy) bis(ethylamine). The authors found that PCL melting transition temperature increased as the shell cross-linked particles core volume increased suggesting that the lamellar thickness of PCL cores in larger shell cross-linked particles are greater. The suppression of the melting peak of the PEO block was also observed for the core cross-linked of PEO-*b*-poly(4-ethynylstyrene) by photoreaction of acetylenic groups.²⁰ The authors of this study considered this result as a fact of the cross-linking caused by UV irradiation. Depressing of the PEO crystallinity in the self-assembly structures of PEO

based block copolymers was largely studied.^{48–51} From the literature, two factors are responsible for this phenomenon: (i) decrease of the PEO segmental mobility caused by the attachment of PEO chain end(s) to a rigid block and (ii) confinement effect due to microphase separation of PEO based block copolymers in nanodomains. Wunder et al. observed in oligomeric PEO- functionalized silsesquioxanes a similar phenomenon that they attributed to the restriction upon chain mobility. In our case, one of the PEO chain ends is anchored to the micelles core, which becomes more and more rigid with increasing of the cross-linking degree. This rigidity may restrict the mobility of the chains and suppress their crystallization (Figure 7b). Interestingly, the crystallinity of the pristine copolymer and their nonirradiated micelles is significantly different even though the latter are not yet core cross-linked (Table 1S, entries 1 and 2). The melting enthalpies of nonirradiated copolymer micelles are approximately 70% of those of the pristine copolymer. In fact, nonirradiated micelles are spherical “particles” with a corona consisting of hydrophilic PEO block and a core consisting of hydrophobic PVBA block. Since water is a very poor solvent for PVBA block, the chains in the core are tightly entangled to minimize the interactions of insoluble block with water; forming that way a compact core of micelles. However, when one chain end of PEO is attached to this core, mobility of PEO block is more affected than that in a free copolymer chain. This result demonstrates one more time the important effect of the chain mobility on thermal behavior of PEO.

When the freeze-drying process is used to dry a semicrystalline polymer solution, Xue et al.⁵² have been demonstrated the concentration dependence of crystallinity of polycarbonate (PC). They found that, in a dilute solution, PC crystallizes more quickly and completely than in a concentrated solution. This observation was attributed to the existence of a concentration boundary in very dilute solution; near the boundary concentration, the solution has a lower chain entanglement level, favoring thus the PC crystallization. Since we used, in this study, the freeze-drying process to recover the cross-linked micelles, it is interesting to know if the crystallization behavior of PEO will be dependent or not of the concentration of the solution from which the sample freeze-dried. For that, DSC measurements were run for two freeze-dried samples of **P1-N₃** copolymer. The first one from

nonirradiated micelles solution at 0.05 wt % and the second one from micelles solution at 0.05 wt % irradiated for 45 min under UV light. As we can note in the obtained results shown in Figure 6S and Table 2S, crystallization temperature of PEO was almost not affected with micelles concentration, on the other hand, the crystallinity degree of PEO was quite different depending micelles solution concentration, particularly when micelles are cross-linked (Table 2S). We observed a decrease of 5% for the degree of crystallinity of cross-linked micelles obtained from dilute micelles solution. This observation is opposite with those observed by Xu et al. for polycarbonate homopolymer of M_w of 30 000 g·mol⁻¹. We explain this by the fact that the studied PEO chains are short (5000 g·mol⁻¹) and near the limit of the entanglement molecular weight,⁵³ which means that there is not influence of chain entanglement on the PEO crystallinity. Therefore, proximity of micelles in a concentrated solution allows PEO chains crystallize easier and higher than distant micelles in a dilute solution.

CONCLUSION

Amphiphilic diblock copolymers consisting of PEO with two different molar masses (with 5000 and 20 000 g/mol) as hydrophilic block and PS derivative bearing azide side group as hydrophobic block were synthesized using NMP technique. Spherical aqueous micelles of 23 and 71 nm have been prepared from these block copolymers. The cores of the obtained micelles have been cross-linked by UV irradiation without addition of photoreagent via the cleavage of azide group and formation of dibenzylidiazene junctions. The cross-linking degree can be easily tuned as a function of UV exposure time. We demonstrated, for the first time, how the thermal behavior of PEO is affected by micellization phenomenon and by the cross-linking of the micelles core. Involvement of PEO block as a part of micelles resulted in a decrease of crystallization temperature and the degree of crystallinity, and that intensified more and more with the core cross-linking degree. Attachment of PEO block to a rigid core modified considerably the thermal behavior of PEO, namely a decrease of about 15 °C for T_c and 50% for X_c of cross-linked micelles submitted to 45 min UV irradiation compared to pristine copolymer. Study of these freeze-dried micelles as organic nanoparticles in PEO based solid electrolyte for lithium battery is under way to improve mechanical properties of materials while preserving their ionic conductivity.

ASSOCIATED CONTENT

Supporting Information

Thermal characteristics of PEO in PEO-*b*-PVBA copolymer micelles before and after being exposed to UV irradiation at different times, evolution of micelle size and particle size distribution of cross-linked micelles at different irradiation times by DLS in water, and influence of micelle solution concentration on the crystallization behavior. This material is available free of charge via the Internet at <http://pubs.acs.org>.

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Notes

The authors declare no competing financial interest.

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