"Sandwich" Microcontact Printing as a Mild Route Towards Monodisperse Janus Particles with Tailored Bifunctionality

Tobias Kaufmann, M. Talha Gokmen, Christian Wendeln, Martin Schneiders, Stefan Rinnen, Heinrich F. Arlinghaus, Stefan A. F. Bon, Filip E. Du Prez, and Bart Jan Ravoo**

Anisotropic particles have properties that are completely different from isotropic particles.[1] With some creativity in the control of the shape[2] and[3]/or[4] chemical composition, an almost infinite collection of anisotropic, "patchy" particles of various sizes can be imagined.[5] One interesting class of patchy particles are Janus particles, named after the ancient Roman god Janus, who was believed to have two opposing faces.[6–8] Much attention is drawn to Janus particles since they may possess amphiphilic character as a result of their two distinct poles or sides, giving rise to improved and even unprecedented performance.[9] Janus particles stabilize (Pickering) emulsions[10] by adsorption at the liquid–liquid interface,[11] move or rotate in response to external stimuli,[12] and self-organize in structures with length scales ranging from nano to macro.[13–15] These unique features of Janus particles have led to novel applications, such as liquid marbles,[16] particle-based displays,[17] bar-coded detection,[18] directionally controlled association with cells,[19] and the catalysis of two distinct reactions in both phases while stabilizing a water–oil emulsion at the interface.[20]

Microfluidics,[17] flow lithography,[3] and electrohydrodynamic co-jetting[19] techniques recently enabled researchers to manufacture Janus particles via the solidification (e.g., polymerization) of two co-flowing solutions. Although they are very efficient and devoid of any postmodification steps, these methods have drawbacks. Firstly, biomolecules and other functional additives may not be compatible with either the solidification process or the precursors and solvents used to prepare the particles. Secondly, the majority of the additives will be buried in the interior of the particles rather than being exposed at the surface. On the other hand, several postmodification techniques have been developed to convert isotropic particles into Janus particles. The most straightforward method is to immobilize particles as a monolayer and to expose the upper faces to metal vapor,[21] etching,[16] UV,[22] or laser irradiation.[23] This process, however, only allows functionalization of one side of the particle. Both sides can be functionalized selectively by using multistep interfacial chemistry, i.e., protection of one face of the particles in one phase (often reversibly gelled) and manipulation of the exposed face, followed by removal of the protecting phase and manipulation of the opposite face of the particles.[13] Although they are quite efficient, these methods are restricted to a limited range of deposits under rather harsh conditions. There is an evident need for a mild and versatile technique to engineer the surface of Janus particles, particularly with biomolecules.

Microcontact printing (μCP) is a soft and efficient additive lithography method utilized to engineer surfaces at the micro- and nanoscale.[24,25] A chemical "ink" is absorbed on a microstructured elastomer (typically poly(dimethyl siloxane) (PDMS)), which acts as a stamp, and transferred to a substrate only in the area of contact. There are only a few reports[26–28] on μCP as a technique to pattern curved surfaces such as colloids. In these studies, electrostatic interactions were exploited to transfer the ink onto the particles through physisorption and typically only one side of the particles was manipulated. A useful extension of μCP is microcontact chemistry, which describes surface patterning via chemical reaction of ink and substrate in the area of contact.[29] The major advantage of microcontact chemistry is that the surface is modified in a permanent and resistant way, i.e., by formation of covalent links between ink and substrate. Recently, a first example of microcontact chemistry on nanoparticles has been demonstrated by printing two fluorescent silanes onto silica nanoparticles.[30]

In this paper we describe a versatile strategy for the preparation of Janus polymer microparticles using a "sandwich" μCP strategy in which the two sides of isotropic spherical particles can be simultaneously modified and distinctively functionalized in a single step. Porous epoxy-functionalized polymer microparticles of monodisperse size distribution were prepared using microfluidics. The polymer microparticles served as a substrate for microcontact chemistry. We made use of an addition reaction between the epoxy group on the polymer particles and functional amines in the ink. This reaction does not require any catalyst and proceeds at moderate temperature (20–120 °C), although both components are stable under ambient conditions. Epoxy-terminated

T. Kaufmann, C. Wendeln, M. Schneiders, Prof. B. J. Ravoo
Organisch-Chemisches Institut
Westfälische Wilhelms-Universität Münster
Corrensstraße 40, 48149 Münster, Germany
E-mail: b.j.ravoo@uni-muenster.de
M. T. Gokmen, Prof. F. E. Du Prez
Department of Organic Chemistry
Ghent University
Krijgslaan 281 S4, 9000 Gent, Belgium
E-mail: Filip.DuPrez@UGent.be
S. Rinnen, Prof. H. F. Arlinghaus
Physikalisches Institut
Westfälische Wilhelms-Universität Münster
Wilhelm-Klemm-Straße 10, 48149 Münster, Germany
Prof. S. A. F. Bon
Department of Chemistry
University of Warwick
Coventry CV4 7AL, UK

DOI: 10.1002/adma.201003564

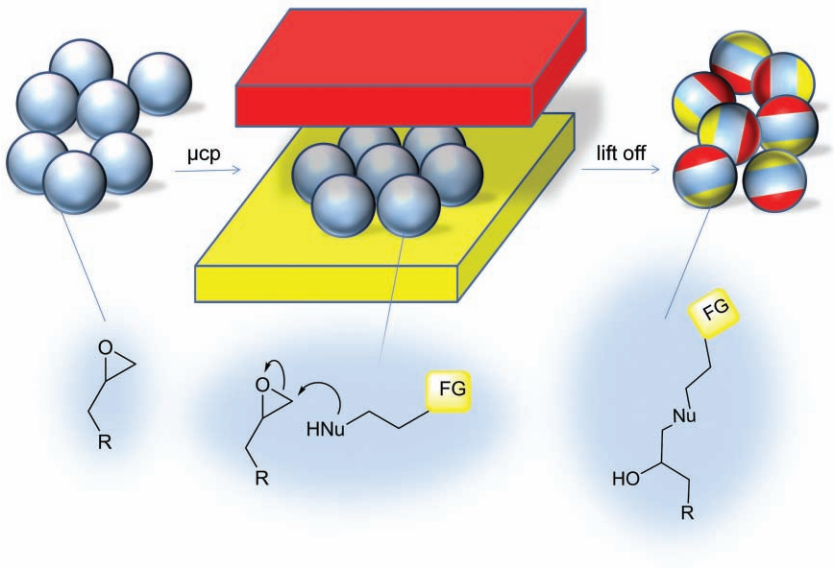

Figure 1. Schematic illustration of the preparation of polymer Janus particles by sandwich microcontact chemistry. Porous particles carrying epoxy moieties made by microfluidics are adsorbed between two stamps loaded with different inks. The inks react with the particles in the area of contact upon heating. Functional Janus particles are obtained after lift-off and extensive rinsing.

self-assembled monolayers (SAMs) on glass and silicon substrates served as references for the reaction on the polymer particles. So far, microcontact chemistry on epoxy surfaces is limited to biotechnological applications, such as the immoblization of proteins on planar surfaces (SAMs).[31,32] The surface reactions by μCP were monitored by optical microscopy, fluorescence microscopy, and time-of-flight secondary ion mass spectrometry (ToF-SIMS).

The approach to prepare bifunctional Janus microparticles is schematically illustrated in **Figure 1**. Porous, monodisperse polymer particles manufactured from ethylene glycol dimethacrylate (EGDMA) and glycidyl methacrylate (GMA) using a microfluidic reactor (particle diameter 160 μm; see Figure S1 in the Supporting Information) were assembled into a monolayer on a flat PDMS stamp loaded with an amine ink. A second flat stamp (loaded with a different amine ink) was placed on top of the particle monolayer. The reaction was induced by heating for 4 h. At room temperature, the amine inks react with the epoxy particles slowly (see Supporting Information) so that if the particles move during the loading of the stamps, they are not significantly derivatized. Next, both stamps were lifted off. The particles were washed extensively to remove nonreacted inks. To show the versatility and scope of the approach, the fabrication of three classes of Janus particles are reported: fluorescent Janus particles made using fluorescent inks, protein-binding Janus particles made using carbohydrate inks, and magnetic Janus particles made using nanoparticle inks.

As a first proof-of-principle experiment, a red and a blue dye (rhodamine ethylenediamine and dansylcadaverine, respectively; see Figure S2 in the Supporting Information) were printed on polymer particles. The printing procedure was as follows: 1) Two flat, oxidized PDMS stamps were incubated with a few drops of the red and blue fluorescent inks (1 mM in ethanol) for 1 min and then dried in a stream of argon; 2) A monolayer of epoxy polymer particles was applied to one stamp while the other stamp was placed on top; 3) The sandwich of stamp-particles-stamp was inserted into a homemade press and heated at 120 °C for 4 h; 4) The stamps were removed and the particles were washed thoroughly with water, ethanol, acetone, and diethylether and subsequently extracted in hot ethanol for 9 h. Fluorescence microscopy images of the resulting Janus particles are shown in **Figure 2**A–C. It is

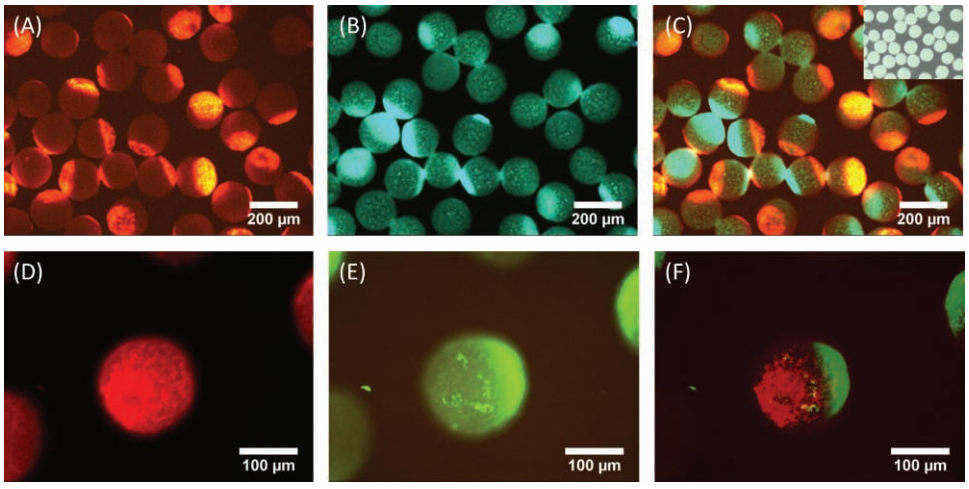

Figure 2. Upper row: Janus polymer particles obtained by printing red and blue dyes: A) rhodamine ethylenediamine (green filter); B) dansylcadaverine (UV filter); and C) overlay of both images. Inset: bright field image. Bottom row: Janus polymer particles obtained by printing carbohydrates: D) rhodamine-labeled PNA bound to β-galactoside-modified faces (green filter); E) Fluorescein-labeled Con A bound to α-mannoside faces (blue filter); and F) overlay of both images.

evident from Figure 2A–C that the functionalization of the particles with the red and blue dyes occurred on the opposing faces of the particles.

A second class of bifunctional Janus particles was equipped with selective biological receptors at each face. To this end, amino-β-galactoside and amino-α-mannoside inks (see Figure S2 in the Supporting Information) were printed on the opposing poles of the epoxy polymer particles. The stamp-particles-stamp sandwich was again heated at 120 °C for 4 h. After printing, the stamps were removed and the particles were washed extensively with water, ethanol, acetone, and diethylether and extracted for 9 h in hot ethanol to remove unreacted carbohydrate ink. The specific carbohydrate–lectin affinities of peanut agglutinin (PNA) to β-galactosides and of concanavalin A (Con A) to α-mannosides were exploited to verify and visualize the Janus character of the carbohydrate functionalized polymer particles (Figure 2D–F). The particles were incubated in a dilute solution of fluorescent PNA and Con A that contained both lectins in equimolar ratio. It is evident from Figure 2D–F that the rhodamine-labeled PNA (red) and fluorescein-labeled Con A (green) adsorbed selectively to opposing faces of the polymer particles. These observations demonstrate that the carbohydrates were immobilized exclusively in the contact areas between the particles and each stamp and that both carbohydrates retain their characteristic affinity for each lectin, also when immobilized on the polymer particles by μCP.

In order to create anisotropic particles that are responsive to a magnetic field, a third set of Janus particles was produced by combining the superparamagnetic properties of magnetite nanoparticles on one face of the epoxy polymer particles with a red fluorescent label (rhodamine ethylenediamine) on the second face (**Figure 3**). In this case one stamp was inked with dopamine-modified, amine-terminated magnetite nanoparticles (see Figure S2, Supporting Information) with a diameter of ≈15 nm by applying a few drops of a 1% suspension of magnetite nanoparticles in water onto a flat, oxidized PDMS stamp and then evaporating the solvent to yield a multilayer of magnetite nanoparticles on the stamp surface. A layer of epoxy polymer microparticles was loaded on the nanoparticle-coated stamp. The other stamp was inked with rhodamine ethylenediamine (see above) and placed on top of the polymer microparticles. The sandwiched particles were heated to 120 °C for 4 h. It can be readily observed in Figure 3A–B that the functionalization of the polymer microparticles with the magnetic nanoparticles and the red dye occurred on the opposing faces of the particles. The images in Figure 3A–B display particles in various orientations. However, the magnetic character of the nanoparticles immobilized on one face of the Janus particles enables the alignment of these particles in an external magnetic field (Figure 3C–D). If the magnet is placed on the right, the nanoparticles are on the right face of the polymer microparticles, while the rhodamine is on the left face. Thus, the magnetic Janus particles display not only a chemical anisotropy but also a magnetic anisotropy, which allows superior control not only over the chemical properties, but also the trajectory and orientation of the particles. The behavior of the magnetic Janus particles is comparable to the well-known effect of magnetic precipitation: the magnetic face of the particle is attracted to the external permanent magnet so that all the Janus particles align in the direction of the magnet.[33,34]

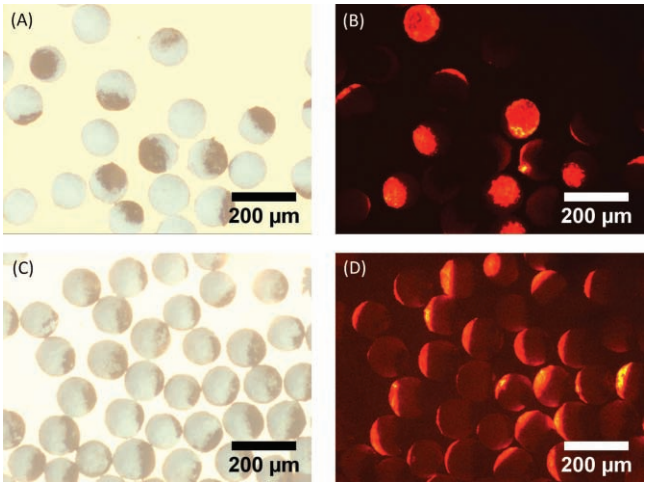

Figure 3. Bright field images (A, C) and fluorescence microscopy images (B, D) of Janus particles reacted with dopamine-coated magnetite nanoparticles on one face and rhodamine ethylenediamine on the opposing face. A ferromagnet was placed on the right side of the particles shown in (C) and (D).

Although it is beyond the scope of this communication to explore the full parameter space of our method, it is clear that the efficacy of sandwich printing will depend on a number of key parameters: a) the modulus of the stamp; b) the modulus of the polymer microspheres; c) the pressure, p, applied in the sandwich press; d) the temperature, T; and e) the particle diameter. In essence, these parameters determine the contact area of the stamp and particle, as well as the kinetics of the immobilization reaction. If the modulus of the stamp or the microparticle is increased, the contact area will be smaller (at constant p, T). If p is increased, the contact area will be larger. If T is increased, the modulus decreases and hence the contact area increases. In addition, an increase in T increases the rate of the immobilization reaction. Concerning the particle size, one expects that for a given combination of modulus, p, and T, the contact area (as a percentage of total area) would be higher for small particles than for large particles. It should be possible to print on much smaller microparticles as well as nanoparticles. Jiang and Granick printed on particles as small as 150 nm in diameter.[30] There could be a lower limit to the particle size due to difficulties in preparing a monolayer of nanoparticles in the stamp sandwich (small nanoparticles tend to form multilayers due to their high surface area) and the rapid diffusion of the ink across the entire surface of such small particles. Also, it is essential that the particles are rather monodisperse, since only in that case both poles can be contacted and reacted with equal efficiency in a sandwich arrangement.

In order to exclude that the preparation of Janus particles by μCP is the result of physisorption or other forms of non-specific adhesion of the inks in the contact area of the stamp and the particles, a number of reference experiments were carried out. First, it was found that μCP on "non-reactive" polymer particles terminated with methyl or butyl ester moieties instead of epoxides did not show significant reactivity towards any amine inks.

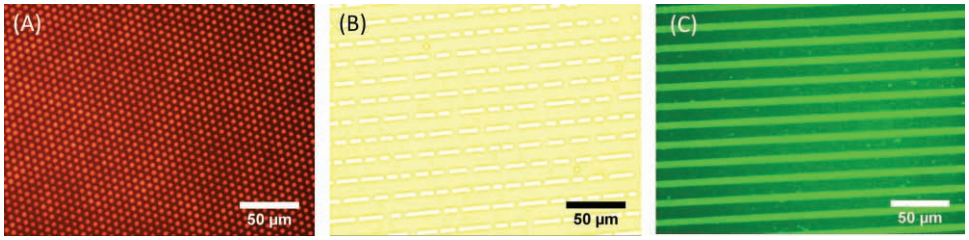

Figure 4. Microcontact chemistry with amine inks on epoxy-terminated SAMs: A) dot pattern of rhodamine ethylenediamine printed on an epoxy-terminated SAM at 120 °C; B) selective water condensation on a line pattern of amino-α-mannoside; and C) fluorescein–labeled Con A selectively bound to the amino-α-mannoside line pattern.

This negative control confirmed that the epoxy group is essential for efficient surface reaction by µCP.

Second, amine inks were printed on epoxy-terminated SAMs to allow for a more detailed surface analysis. In this case, the reaction in the contact area of a micropatterned PDMS stamp and an epoxy-terminated SAM serves as a reference for the reaction in the contact area of a flat PDMS stamp and a polymer particle. Rhodamine ethylenediamine, amino-α-mannoside, and amino-β-galactoside were used as inks for µCP (**Figure 4** and Supporting Information). It can be seen that rhodamine ethylenediamine can be printed efficiently at an epoxy-terminated SAM (Figure 4A). It was found that the reaction by µCP is also possible at room temperature, although with significantly lower immobilization density (see Supporting Information, Figure S3). In addition, patterns of amino-α-mannoside were printed on epoxy-terminated SAMs. The patterns could be visualized by selective water condensation due to the higher hydrophilicity of the carbohydrate areas compared to the epoxy substrate (Figure 4B). Moreover, the patterns could be visualized by incubation in fluorescein–labeled Con A, which resulted in a selective protein binding on the carbohydrate areas (Figure 4C). Finally, amino-β-galactoside could be printed on an epoxy-terminated SAM (see Figure S4,S5 in the Supporting Information).

In conclusion, we have shown that covalently bifunctionalized polymer Janus particles can be prepared by a sandwich microcontact chemistry strategy. Janus particles with distinct optical, biological, and magnetic functions on opposing faces could be obtained using this approach. Since the Janus particles are porous, they can potentially be filled with guest materials, including catalysts.[34] We believe that this method is versatile and potent and allows for the preparation of functional Janus particles under mild conditions.

Experimental Section

Preparation of porous particles: Typically, particles were prepared by using a tubing-needle-based microfluidic setup.[35] Tygon tubing (2 m length, 0.8 mm inner diameter) served as the channel and 30G or 32G bent blunt needles were used for the discrete monomer phase. Pumping rates were 1.2 mL min^{-1} and 0.60 mL h^{-1} for the continuous and discrete phases, respectively. The continuous carrier phase was a 3 wt% SDS solution and the discrete phase was composed of 20 vol% EGDMA (crosslinker), 30 vol% GMA (epoxy monomer), 50 vol% *n*-octanol (porogen), and 4 wt% (compared to EGDMA+GMA) 2,2-dimethoxy-2-phenylacetophenone (DMPA, photoinitiator). Details about the various porogens (important for "skin"[36] formation) and photoinitiators (important for the inherent fluorescence of particles) studied can be found in the Supporting Information.

Microcontact printing on particles: Stamps were made from PDMS (see Supporting Information). Oxidized stamps were loaded with a few drops of ink solution (1–10 mM in ethanol with triethylamine added as a base) for 1 min. Excess ink solution was removed under a stream of argon for approximately 1 min. A monolayer of epoxy polymer particles was applied to one stamp while the other stamp was placed on top. The sandwich of stamp-particles-stamp was inserted into a homemade hand-operated press and heated at 120 °C for 4 h. When the reaction was complete, the polymer particles were collected in a frit and washed with water, ethanol, acetone, and diethylether. Samples were extracted in hot ethanol for 9 h.

Supporting Information

Supporting Information is available from the Wiley Online Library or from the author.

Acknowledgements

This work was supported by a Marie Curie Early Stage Research Training Fellowship of the European Community's Sixth Framework Program under contract number 020643 (M.T.G, Sendichem project) and the Belgian Program on Interuniversity Attraction Poles initiated by the Belgian State, Prime Minister's office (Program P6/27).

Received: September 29, 2010
Published online: November 11, 2010

[1] D. Dendukuri, P. S. Doyle, *Adv. Mater.* **2009**, *21*, 4071.
[2] D. C. Pregibon, M. Toner, P. S. Doyle, *Science* **2007**, *315*, 1393.
[3] K. W. Bong, K. T. Bong, D. C. Pregibon, P. S. Doyle, *Angew. Chem. Int. Ed.* **2010**, *49*, 87.
[4] H. Takeshi, T. Atsunori, M. Kiwamu, Y. Hiroshi, S. Masatsugu, *Angew. Chem. Int. Ed.* **2008**, *47*, 8044.
[5] S. C. Glotzer, M. J. Solomon, *Nat. Mater.* **2007**, *6*, 557.
[6] A. Perro, S. Reculusa, S. Ravaine, E. B. Bourgeat-Lami, E. Duguet, *J. Mater. Chem.* **2005**, *15*, 3745.
[7] F. Wurm, A. F. M. Kilbinger, *Angew. Chem. Int. Ed.* **2009**, *48*, 8412.
[8] A. Walther, A. H. E. Müller, *Soft Matter* **2008**, *4*, 663.
[9] S. Jiang, Q. Chen, M. Tripathy, E. Luijten, K. S. Schweizer, S. Granick, *Adv. Mater.* **2010**, *22*, 1060.
[10] J.-W. Kim, D. Lee, H. C. Shum, D. A. Weitz, *Adv. Mater.* **2008**, *20*, 3239.
[11] D. L. Cheung, S. A. F. Bon, *Soft Matter* **2009**, *5*, 3969.
[12] C. H. Chen, A. R. Abate, D. Y. Lee, E. M. Terentjev, D. A. Weitz, *Adv. Mater.* **2009**, *21*, 3201.
[13] X. Y. Ling, I. Y. Phang, C. Acikgoz, M. D. Yilmaz, M. A. Hempenius, G. J. Vancso, J. Huskens, *Angew. Chem. Int. Ed.* **2009**, *48*, 7677.
[14] S. Whitelam, S. A. F. Bon, *J. Chem. Phys.* **2010**, *132*, 074901.

[15] W. L. Miller, A. Cacciuto, *Phys. Rev. E* **2009**, *80*, 021404.
[16] S.-H. Kim, S. Y. Lee, S.-M. Yang, *Angew. Chem. Int. Ed.* **2010**, *49*, 2535.
[17] T. Nisisako, T. Torii, T. Takahashi, Y. Takizawa, *Adv. Mater.* **2006**, *18*, 1152.
[18] K. W. Bong, S. C. Chapin, P. S. Doyle, *Langmuir* **2010**, *26*, 8008.
[19] M. Yoshida, K.-H. Roh, S. Mandal, S. Bhaskar, D. Lim, H. Nandivada, X. Deng, J. Lahann, *Adv. Mater.* **2009**, *21*, 4920.
[20] S. Crossley, J. Faria, M. Shen, D. E. Resasco, *Science* **2010**, *327*, 68.
[21] S. K. Smoukov, S. Gangwal, M. Marquez, O. D. Velev, *Soft Matter* **2009**, *5*, 1285.
[22] H.-Y. Chen, J.-M. Rouillard, E. Gulari, J. Lahann, *Proc. Natl. Acad. Sci.* **2007**, *104*, 11173.
[23] E. Hugonnot, A. Carles, M. H. Delville, P. Panizza, J. P. Delville, *Langmuir* **2003**, *19*, 226.
[24] S. A. Ruiz, C. S. Chen, *Soft Matter* **2007**, *3*, 168.
[25] T. Kaufmann, B. J. Ravoo, *Polym. Chem.* **2010**, *1*, 371.
[26] Z. Li, D. Lee, M. F. Rubner, R. E. Cohen, *Macromolecules* **2005**, *38*, 7876.
[27] O. Cayre, V. N. Paunov, O. D. Velev, *J. Mater. Chem.* **2003**, *13*, 2445.
[28] O. Cayre, V. N. Paunov, O. D. Velev, *Chem. Commun.* **2003**, 2296.
[29] B. J. Ravoo, *J. Mater. Chem.* **2009**, *19*, 8902.
[30] S. Jiang, S. Granick, *Langmuir* **2009**, *25*, 8915.
[31] B. Thierry, M. Jasieniak, L. C. P. M. de Smet, K. Vasilev, H. J. Griesser, *Langmuir* **2008**, *24*, 10187.
[32] J.-H. Lee, H.-W. Shim, H.-S. Choi, Y.-A. Son, C.-S. Lee, *J. Phys. Chem. Solids* **2008**, *69*, 1581.
[33] N. Prasad, J. Perumal, C.-H. Choi, C.-S. Lee, D.-P. Kim, *Adv. Funct. Mater.* **2009**, *19*, 1656.
[34] C. P. Park, D.-P. Kim, *Angew. Chem. Int. Ed.* **2010**, *49*, 6825.
[35] M. T. Gokmen, W. Van Camp, P. J. Colver, S. A. F. Bon, F. E. Du Prez, *Macromolecules* **2009**, *42*, 9289.
[36] S. Dubinsky, J. I. Park, I. Gourevich, C. Chan, M. Deetz, E. Kumacheva, *Macromolecules* **2009**, *42*, 1990.