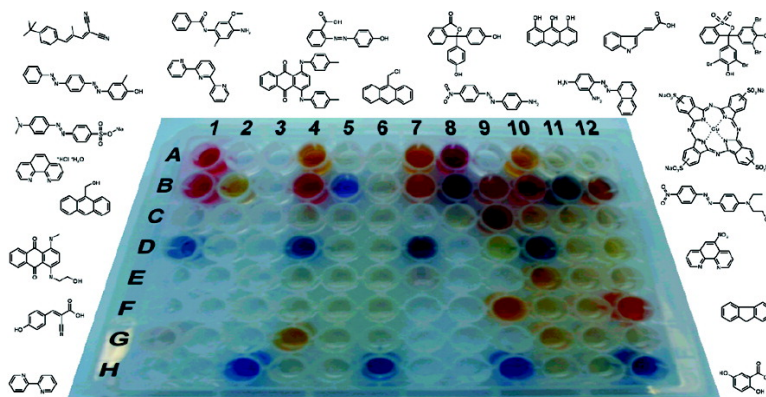


## Combinatorial Evaluation of the Host–Guest Chemistry of Star-Shaped Block Copolymers

Michael A. R. Meier, and Ulrich S. Schubert

*J. Comb. Chem.*, **2005**, 7 (3), 356-359 • DOI: 10.1021/cc049824t • Publication Date (Web): 12 April 2005

Downloaded from <http://pubs.acs.org> on March 22, 2009



### More About This Article

Additional resources and features associated with this article are available within the HTML version:

- Supporting Information
- Links to the 3 articles that cite this article, as of the time of this article download
- Access to high resolution figures
- Links to articles and content related to this article
- Copyright permission to reproduce figures and/or text from this article

[View the Full Text HTML](#)

## Combinatorial Evaluation of the Host–Guest Chemistry of Star-Shaped Block Copolymers

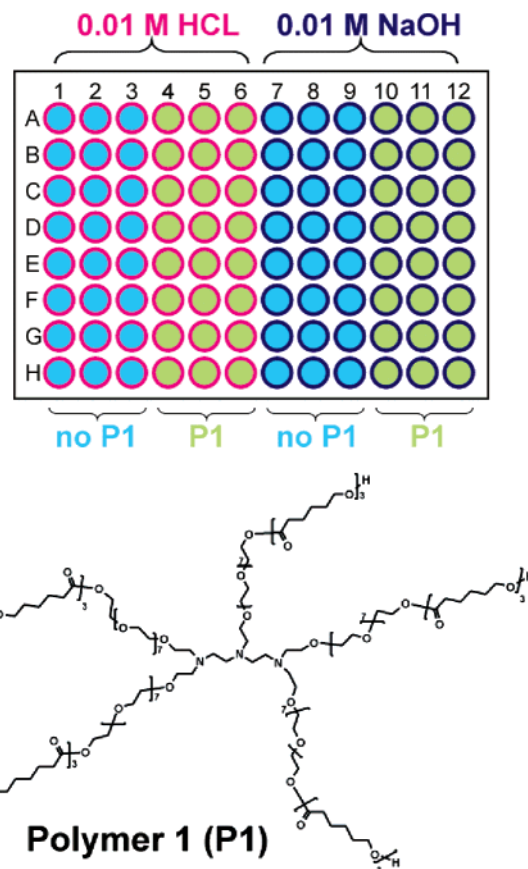
Michael A. R. Meier and Ulrich S. Schubert\*

Laboratory of Macromolecular Chemistry and Nanoscience, Eindhoven University of Technology and Dutch Polymer Institute (DPI), PO Box 513, 5600 MB Eindhoven, The Netherlands

Received November 9, 2004

The encapsulation of guest molecules in unimolecular micelles based on dendrimers or branched macromolecules as host systems is an interesting field of research, since the resulting supramolecular host–guest systems might be suitable for the controlled delivery of drugs,<sup>1–4</sup> micellar catalysis,<sup>5,6</sup> or sensoric applications.<sup>7</sup> Advantages of drug encapsulation with dendritic macromolecules are, for instance, the solubilization of nonsoluble drugs,<sup>8,9</sup> which is one of the current problems with the delivery of low molecular weight drugs,<sup>10</sup> or the reduction of toxic effects.<sup>11</sup> On the other hand, it was shown that reversed unimolecular micelles based on dendritic systems could act as nanoscale catalytic systems for organic reactions with a polar transition state due to their defined nanoenvironment.<sup>5</sup> Recently, we have shown that star-shaped block copolymers consisting of a poly(ethylene glycol) core and a poly( $\epsilon$ -caprolactone) corona behave as unimolecular micelles that can encapsulate and transfer methyl orange from a water to a chloroform phase.<sup>12</sup> Within this contribution, we describe a fast screening method for the evaluation of the host–guest chemistry of the mentioned polymer class with 24 different UV absorbing guest molecules as well as a few potential applications of these polymers.

The encapsulation of guest molecules within a host can be probed by the evaluation of changes in their microenvironment with techniques such as UV/vis and fluorescence spectroscopy, NMR spectroscopy, or others.<sup>13</sup> These changes are premised on stabilizing or destabilizing effects of the local microenvironment on electronic states of the guest molecules. Our primary and fast screening of host–guest interactions of 24 organic molecules with the above-mentioned star-shaped block copolymers was based on an evaluation of the changes in the molecules' microenvironments by UV/vis as well as fluorescence spectroscopy. Therefore, a microtiter plate screening assay (Figure 1, top for the layout) in combination with a plate reader that allows the fast recording of full UV/vis and fluorescence spectra in a quasi parallel mode was utilized. This setup allowed recording of all necessary UV/vis as well as fluorescence spectra for the screening of a 96-well microtiter plate in less than 15 min (measuring 96 UV/vis spectra could be performed in  $\sim 40$  s, whereas the recording of 96 fluorescence spectra took roughly 2 min). The investigated unimolecular micelles<sup>12</sup> for this screening approach were star-shaped block copolymers consisting of a poly(ethylene glycol) core and a poly( $\epsilon$ -caprolactone) corona, which was prepared in larger scale (55 g,  $M_n = 3350$ , PDI = 1.21; Figure 1, right).<sup>14</sup> The poly( $\epsilon$ -caprolactone) corona was chosen with only three average repeat units to provide sufficient water solubility of

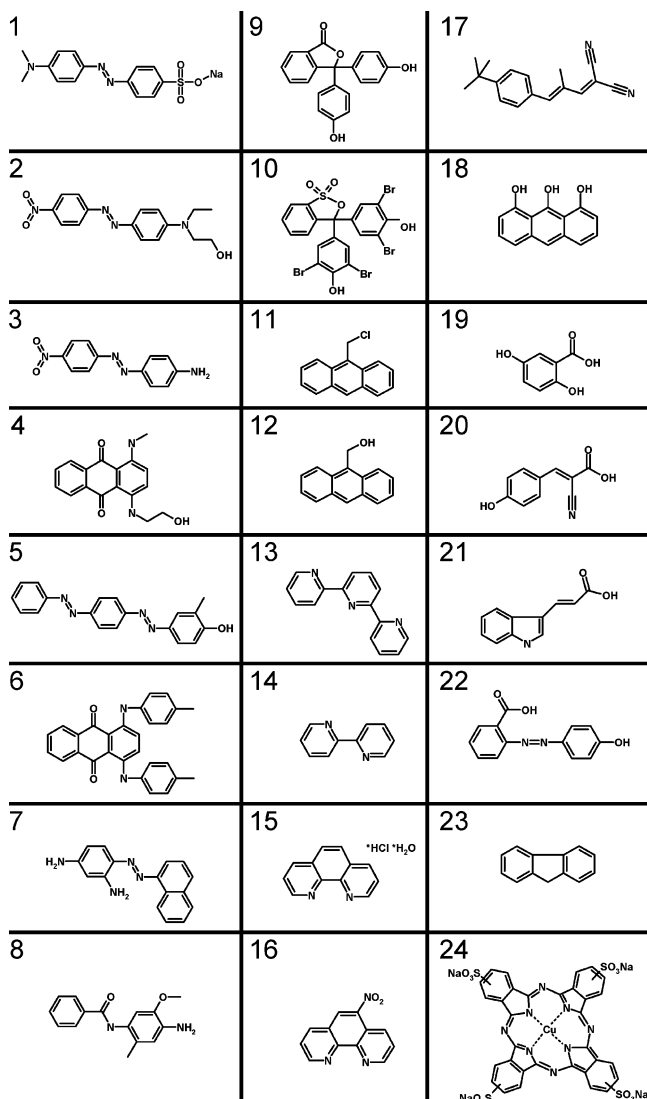


**Figure 1.** A schematic representation of the applied layout for the screening (top) as well as the structure of the utilized star-shaped block copolymer (please note that the repeat unit number of the PEG and PCL blocks are averaged numbers).

the polymer (the size of the corona was found to have no influence on the host–guest properties of the discussed polymers, as described recently<sup>12</sup>). To show that the investigated polymer **P1** exists as a unimolecularly dissolved species not only in tetrahydrofuran and chloroform as described recently<sup>12</sup> but also in an aqueous environment, dynamic light scattering experiments in water were performed. One could expect the formation of larger aggregates of polymer molecules with the poly(ethylene glycol) part on the outer surfaces and the more lipophilic poly( $\epsilon$ -caprolactone) part in the inside of the aggregate due to the different solubilities of the two block components in water. However, the performed light-scattering experiments did not reveal any aggregates of copolymer **P1** in a concentration range from 0.001 to 100 mg/mL leading to the conclusion that the observed effects can be attributed to a unimolecular behavior even if the absolute conformation of these macromolecules is considerably different in an aqueous solution.

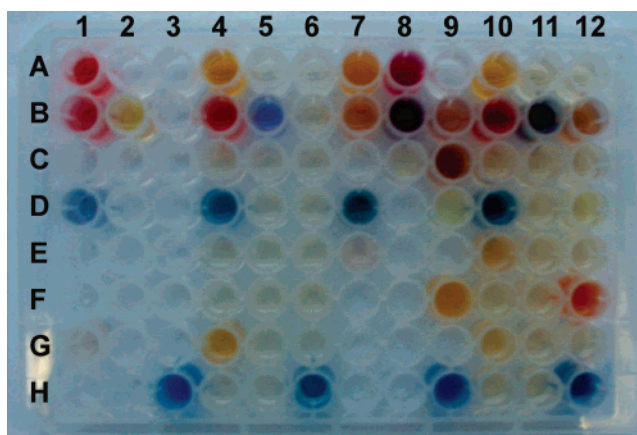
The screening was performed in acidic as well as basic aqueous environments to ensure that changes in the microenvironment of the investigated guest molecules could be detected even for pH-active guest molecules. A more detailed discussion of the necessity to utilize different environments for this screening approach is given below. Both acidic and basic environments could lead to a hydrolysis of the ester bonds of the PCL block of polymer **P1**; however, no change

\* Corresponding author. E-mail: u.s.schubert@tue.nl

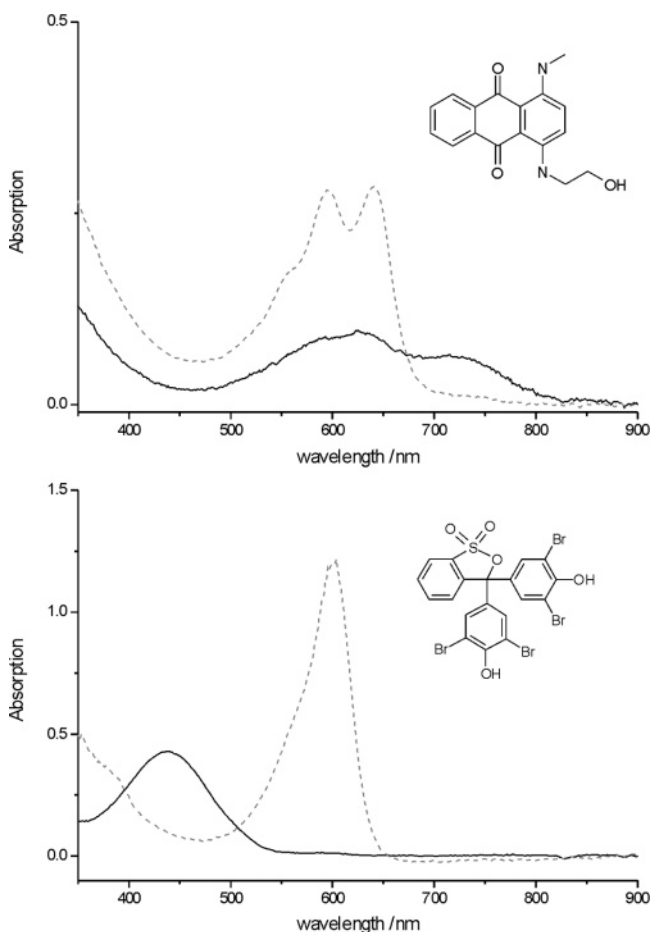


**Figure 2.** Chemical structures as well as digit code of all investigated potential guest molecules.

in the molecular weight distribution of **P1** could be observed after storage in 0.01 M HCl or 0.01 M NaOH for 24 h by gel permeation chromatography. This time is well above the time required to perform all necessary measurements for this screening. Nevertheless, it would be interesting to study the dependence of the degradation behavior of this class of polymers on pH and other factors in the future. Figure 2 provides the chemical structures of all investigated potential guest molecules together with a digit code for straightforward compound identification (see Supporting Information for the chemical names of the investigated guest molecules). The plate was filled with 20  $\mu\text{L}$  equiv of saturated solutions of all 24 guest molecules in 0.01 M HCl (positions A1–H6) and 0.01 M NaOH (positions A7–H12), respectively (see color coding in Figure 1, top). Subsequently, the solutions of guest molecules were diluted to 250  $\mu\text{L}$  with 0.01 M HCl (positions A1–H3), 0.01 M NaOH (positions A7–H9), 200 mg/mL polymer **P1** in 0.01 M HCl (positions A4–H6), and 200 mg/mL polymer **P1** in 0.01 M NaOH (positions A10–H12), respectively. This resulted in four sets of the 24 guest molecules, explicitly guest only and guest in the presence of polymer **P1** in both acidic and basic environmenta. UV/vis spectra in a range from 250 to 900 nm as well as fluorescence spectra with different excitation wavelengths (390–490 nm in 20-nm steps) were recorded utilizing the described plate



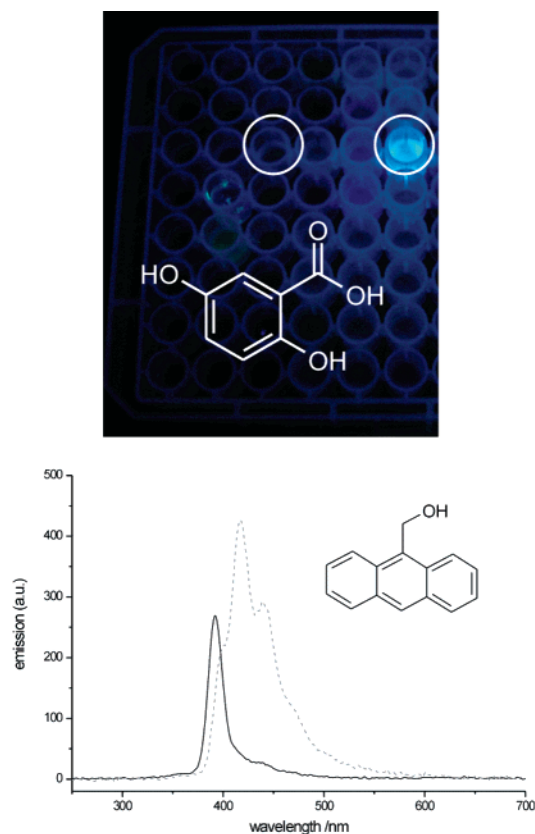
**Figure 3.** Optical picture of the microtiter plate used to screen for host–guest interactions.



**Figure 4.** Encapsulation behavior of guest **4** (positions D7 and D10 in the microtiter plate; compare to Figure 1) in 0.01 M NaOH (top) and guest **10** (positions B8 and B11 in the microtiter plate; compare to Figure 1) in 0.01 M HCl (bottom) visualized by UV/vis spectroscopy; (---) encapsulated, (—) not encapsulated.

reader setup. The measurement of all 96 full UV/vis spectra could be performed in less than 40 s, whereas the measurement of 96 fluorescence spectra took  $\sim 2$  min. Moreover, the plate was inspected visually in daylight as well as under 254- and 365-nm UV irradiation. The screening was completed by diluting the plate by a 1:10 factor, since some of the measured spectra showed high UV/vis absorption. Moreover, the screening was repeated in dichloromethane to avoid any solubility problems of the guest molecules in the aqueous medium and to investigate the encapsulation of

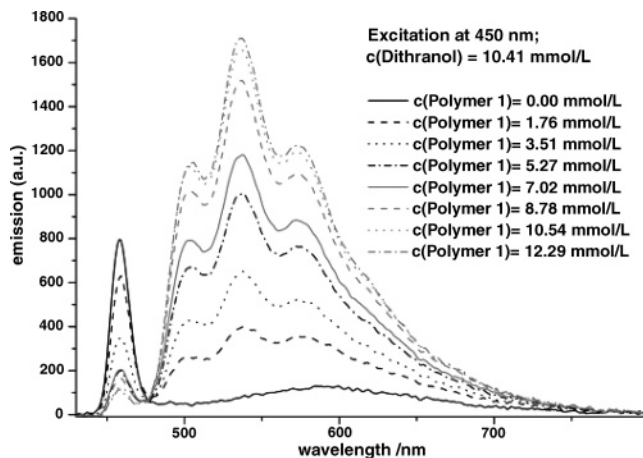




**Figure 5.** Fluorescence intensity increase due to encapsulation of two guest molecules. Top: optical picture under 365-nm irradiation taken of guest **19** in acidic environment (positions C3 and C6 in the microtiter plate; compare to Figure 1). Bottom: fluorescence of guest **12** under 390-nm excitation in 0.01 M NaOH (positions D8 and D11 in the microtiter plate; compare to Figure 1); (---) encapsulated, (—) not encapsulated.

guest molecules from an organic environment. Subsequently, the recorded spectra were utilized to evaluate significant changes in the UV/vis or fluorescence behavior of the 24 investigated guest molecules. The following discussion will only provide screening data of selected guest molecules, since the discussion of all 24 molecules in detail would be too complex and excessively long. The final assessment of all 24 screened molecules as well as additional screening data is provided in the Supporting Information.

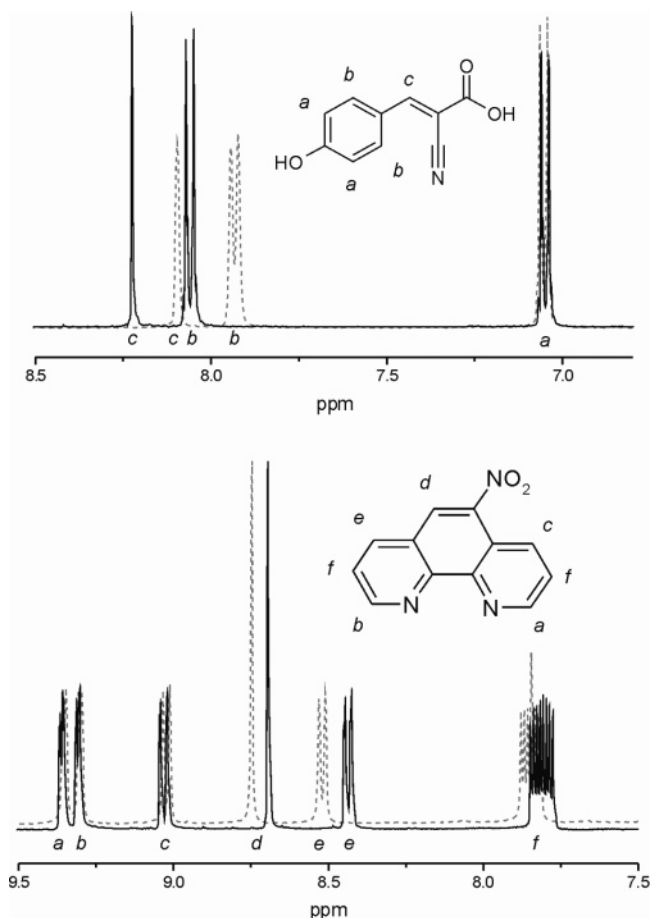
Figure 3 displays an optical picture of the filled microtiter plate as it was utilized for the screening. Already an optical investigation can clearly reveal that some of the investigated guest molecules show an interaction with the star-shaped block copolymer. For instance the guest molecule **9** (phenolphthalein, a pH indicator) in positions A2 (acidic environment), A5 (acidic environment in the presence of polymer **P1**), A8 (basic environment) and A11 (basic environment in the presence of polymer **P1**) clearly shows a color change from red to colorless upon addition of polymer **P1** in the basic environment, but not in the acidic medium. This can be explained by an encapsulation of phenolphthalein and, therefore, a change in the microenvironment of the dyes. However, since phenolphthalein is colorless in acidic solution, this effect is only visible in the basic environment, where this pH indicator shows a red color. The example of phenolphthalein encapsulation also clearly demonstrates the need to screen for host–guest interactions in both acidic and basic solutions, since otherwise, some hits might not be identified in the screening procedure. Generally, it was



**Figure 6.** Polymer **P1** concentration-dependent fluorescence increase of guest **18** (dithranol) due to its encapsulation in dichloromethane.

observed for a number of guests that the capability of being encapsulated by polymer **P1** was independent of the environment (e.g., solvent or pH) where the encapsulation took place. However, the environment was important for the detection of the encapsulation, as described for the case of phenolphthalein, and therefore, different environments (solvent and pH) were chosen for this screening approach for a complementary assessment of the host–guest chemistry of the investigated reversed unimolecular micelles.

A careful investigation of the recorded UV/vis absorption spectra revealed that 7 of the 24 molecules in HCl and 12 of the 24 molecules in NaOH showed a distinct shift (>25 nm) of the absorption maximum upon addition of the star-shaped block copolymer. This can be interpreted as encapsulation and is recorded as a hit in the sense of the screening. Figure 4 shows UV/vis spectra of guest **4** (disperse blue 3, top) and guest **10** (bromphenolblue, bottom) as examples of this behavior. Both spectra reveal a pronounced shift of the absorption maximum due to a change in the microenvironment of these guest molecules upon addition of the host polymer, which acts as a unimolecular micelle and encapsulates the guest molecules. Moreover, the screening for changes in the fluorescence behavior upon encapsulation of guest molecules revealed that 5 of the 24 guest molecules show a distinct fluorescence increase upon addition of star-shaped block copolymer **P1**. Figure 5 demonstrates this effect for guests **12** (9-(hydroxymethyl)anthracene, bottom) and **19** (2,5-dihydroxybenzoic acid, top) in the presence and absence of the star-shaped block copolymer **P1**. This might be explained by a reduction of self-quenching due to isolation of these guest molecules upon encapsulation and is an interesting property for sensoric applications.<sup>7</sup> Moreover, it was observed that the described changes in UV/vis and fluorescence behavior depend on both guest and polymer concentration. One example of this behavior is provided in Figure 6, where different amounts of polymer **P1** are added to a 10.41 mM solution of guest **18** (dithranol) in dichloromethane. Upon addition of the polymer, more and more of the dithranol guest molecules are encapsulated and, therefore, show a fluorescence increase due to reasons described above. A similar increase was observed if different amounts of dithranol were added to a polymer solution. These concentration-dependent effects were observed not only for dithranol but also for other guest molecules and indicate that the investigated polymer class might be a useful material for the development of optical sensors.



**Figure 7.**  $^1\text{H}$  NMR investigations of guests **16** in  $\text{CDCl}_3$  (bottom) and **20** in acetone- $d_6$  (top) in the absence and presence of polymer **P1**; (---) encapsulated, (—) not encapsulated.

The complete screening of all 24 molecules by UV/vis and fluorescence spectroscopy in acidic, basic, and organic environments revealed 17 successful encapsulations. However, to make sure that all hits were correctly identified and no hits were omitted,  $^1\text{H}$  NMR investigations in different solvents were performed after the screening was finished. Figure 7 shows  $^1\text{H}$  NMR spectra of guest **16** (5-nitro-1,10-phenanthroline, bottom) and guest **20** ( $\alpha$ -cyano-4-hydroxycinnamic acid, top) in the encapsulated and the not encapsulated forms with the corresponding assigned signals in the aromatic region of the spectrum. A change in the microenvironment and, therefore, an encapsulation of the guests can once more explain the observed shifts of the aromatic protons of guests **16** and **20**. Moreover, it was observed that polymer **P1** had a large solubilizing effect on many guests in solvents such as acetone- $d_6$ ,  $\text{CDCl}_3$ , or  $\text{D}_2\text{O}$ , which allows an easy characterization of insoluble molecules in parts-per-million ranges of 4.5 and above if the molecules are encapsulated.

In summary, the  $^1\text{H}$  NMR investigations could identify five more successful encapsulations, resulting in an overall result of 22 hits out of 24 screened molecules (see Supporting Information). Therefore, the primary screening was able to identify 77% of all successful encapsulations and did not identify any false positives. The fact that only 2 of the 24 investigated guests did not show any encapsulation behavior

makes the investigated polymer class a highly promising candidate for drug delivery applications. Moreover, it was observed that the investigated star-shaped block copolymer had a large solubilizing effect on many of the investigated guests in a variety of solvents. This is another interesting feature of these polymers with regard to drug delivery, since nonsolubility of drugs can be a major problem when developing a new therapy.

In conclusion, these findings clearly demonstrate that combinatorial materials research<sup>15</sup> can significantly contribute to a large number of fields of research and can help to reduce the time and effort required for the evaluation of important material properties. Moreover, we were able to show that star-shaped block copolymers consisting of a poly(ethylene glycol) core and a poly( $\epsilon$ -caprolactone) corona are multifunctional materials with a large potential for interesting applications, such as the development of optical sensors.

**Acknowledgment.** The authors thank the Dutch Polymer Institute (DPI) and the Fonds der chemischen Industrie for their financial support. The authors express their sincere thanks to Dr. Bas G. G. Lohmeijer for assistance with the light-scattering experiments.

**Supporting Information Available.** Supporting Information as described in text is available. This material is available free of charge via the Internet at <http://pubs.acs.org>.

## References and Notes

- Boas, U.; Heegaard, P. M. H. *Chem. Soc. Rev.* **2004**, *33*, 43–63.
- Moorefield, C. N.; Newkome, G. R. *C. R. Chimie* **2003**, *6*, 715–724.
- Haag, R. *Angew. Chem., Int. Ed.* **2004**, *43*, 278–282.
- Liu, M.; Kono, K.; Fréchet, J. M. J. *J. Control. Release* **2000**, *65*, 121–131.
- Piotti, M. E.; Rivera, F., Jr.; Bond, R.; Hawker, C. J.; Fréchet, J. M. F. *J. Am. Chem. Soc.* **1999**, *121*, 9471–9472.
- Astruc, D.; Chardac, F. *Chem. Rev.* **2001**, *101*, 2991–3023.
- Fernandez, Y. D.; Gramatges, A. P.; Amendola, V.; Foti, F.; Mangano, C.; Pallavicini, P.; Patronib, S. *Chem. Commun.* **2004**, *14*, 1650–1651.
- Morgan, M. T.; Carnahan, M. A.; Immoos, C. E.; Ribeiro, A. A.; Finkelstein, S.; Lee, S. J.; Grinstaff, M. W. *J. Am. Chem. Soc.* **2003**, *125*, 15485–15489.
- Benito, J. M.; Gómez-García, M.; Ortiz Mellet, C.; Bausanne, I.; Defaye, J.; García Fernández, J. M. *J. Am. Chem. Soc.* **2004**, *126*, 10355–10363.
- Breimer, D. D. *J. Control. Release* **1999**, *62*, 3–6.
- Neerman, M. F.; Chen, H.-T.; Parrish, A. R.; Simanek, E. E. *Mol. Pharm.* **2004**, *1*, 390–393.
- Meier, M. A. R.; Gohy, J.-F.; Fustin, C.-A.; Schubert, U. S. *J. Am. Chem. Soc.* **2004**, *126*, 11517–11521.
- Hecht, S.; Fréchet, J. M. J. *Angew. Chem., Int. Ed.* **2001**, *40*, 74–91.
- The synthesis was performed as described in ref 12 on lab scale with 25 mL (25.8 g, 225.6 mmol) of  $\epsilon$ -caprolactone, 32.3 g (15.0 mmol) of 5-arm poly(ethylene glycol) macro-initiator, and 1/20th of stannous octoate catalyst at 130 °C for 6 h.
- Meier, M. A. R.; Schubert, U. S. *J. Mater. Chem.* **2004**, *14*, 3289–3299.