

# DNA Block Copolymers

Schnitzler, Tobias; Herrmann, Andreas

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# DNA Block Copolymers: Functional Materials for Nanoscience and Biomedicine

TOBIAS SCHNITZLER AND ANDREAS HERRMANN*

*Zernike Institute for Advanced Materials, University of Groningen, Groningen, The Netherlands*

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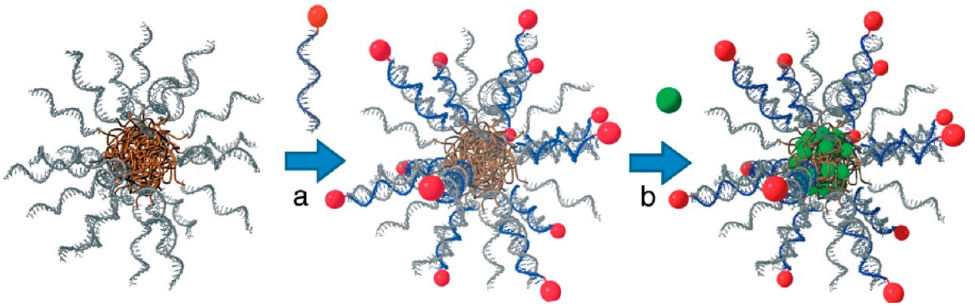
## CONSPECTUS




We live in a world full of synthetic materials, and the development of new technologies builds on the design and synthesis of new chemical structures, such as polymers. Synthetic macromolecules have changed the world and currently play a major role in all aspects of daily life. Due to their tailorable properties, these materials have fueled the invention of new techniques and goods, from the yogurt cup to the car seat belts. To fulfill the requirements of modern life, polymers and their composites have become increasingly complex. One strategy for altering polymer properties is to combine different polymer segments within one polymer, known as block copolymers. The microphase separation of the individual polymer components and the resulting formation of well defined nanosized domains provide a broad range of new materials with various properties. Block copolymers facilitated the development of innovative concepts in the fields of drug delivery, nanomedicine, organic electronics, and nanoscience.

Block copolymers consist exclusively of organic polymers, but researchers are increasingly interested in materials that combine synthetic materials and biomacromolecules. Although many researchers have explored the combination of proteins with organic polymers, far fewer investigations have explored nucleic acid/polymer hybrids, known as DNA block copolymers (DBCs). DNA as a polymer block provides several advantages over other biopolymers. The availability of automated synthesis offers DNA segments with nucleotide precision, which facilitates the fabrication of hybrid materials with monodisperse biopolymer blocks. The directed functionalization of modified single-stranded DNA by Watson–Crick base-pairing is another key feature of DNA block copolymers. Furthermore, the appropriate selection of DNA sequence and organic polymer gives control over the material properties and their self-assembly into supramolecular structures. The introduction of a hydrophobic polymer into DBCs in aqueous solution leads to amphiphilic micellar structures with a hydrophobic polymer core and a DNA corona.

In this Account, we discuss selected examples of recent developments in the synthesis, structure manipulation and applications of DBCs. We present achievements in synthesis of DBCs and their amplification based on molecular biology techniques. We also focus on concepts involving supramolecular assemblies and the change of morphological properties by mild stimuli. Finally, we discuss future applications of DBCs. DBC micelles have served as drug-delivery vehicles, as scaffolds for chemical reactions, and as templates for the self-assembly of virus capsids. In nanoelectronics, DNA polymer hybrids can facilitate size selection and directed deposition of single-walled carbon nanotubes in field effect transistor (FET) devices.

## Introduction

Block copolymers and their supramolecular assemblies have attained much attention since their structural properties, either in solid state or in solution, became accurately predictable.[1−3] The nanometer-sized superstructures formed by block copolymers depend on the chemical nature of the monomers, the length of the individual blocks, and the block length ratio. For the adjustment of ordered domains, it is crucial to control the molecular parameters of the individual block copolymer chains. This holds true for block

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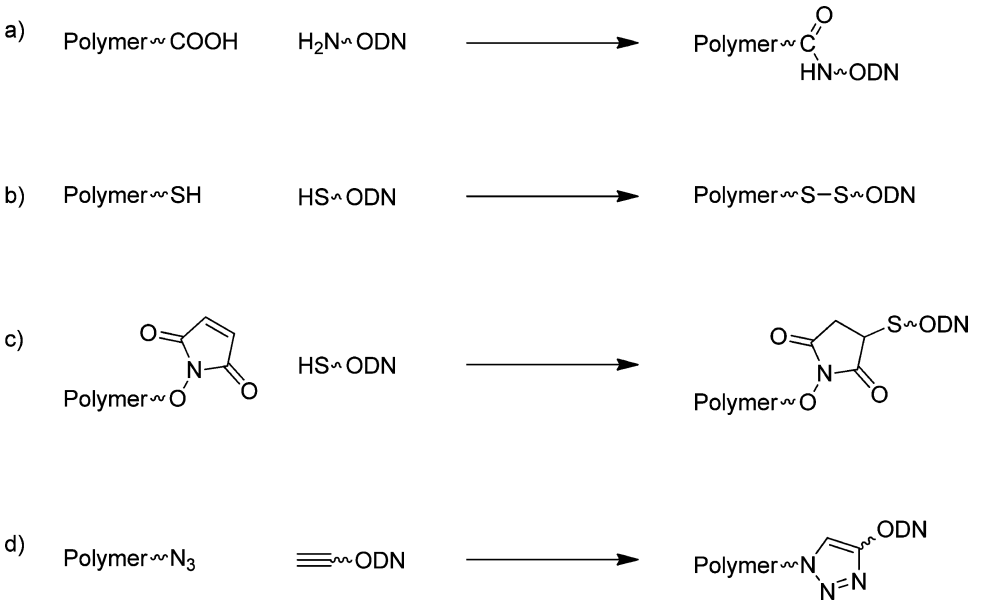
**SCHEME 1.** Coupling Methods for Solution-Based DBC Synthesis[a]




[a](a) Carboxylic acid-terminated polymer coupled with an amino-functionalized ODN. (b) Disulfide bond formation between polymer and ODN thiol. (c) Michael addition of a terminal maleimide on the polymer and thiol-modified ODN. (d) Click reaction between an azide-functionalized polymer and an alkyne-modified ODN.

copolymers consisting exclusively of organic monomers as well as for block polymer architectures containing biomacromolecule segments. In this respect, peptide organic polymer conjugates are an established and well investigated type of hybrid structures.[4−8] Much less attention has been paid to the class of bioorganic hybrids consisting of nucleic acids and synthetic polymers.[9,10]

The first reports of DNA−polymer hybrid materials date back to the late 1980s. By grafting antisense oligodeoxyribonucleotides (ODNs) onto a poly(L-lysine) (PLL) backbone, antiviral agents were formed that inhibited the synthesis of vesicular stomatitis virus.[11,12] Since then, a number of applications have been realized with this class of materials in addition to gene or ODN delivery. These range from purification of biomaterials[13] to DNA detection.[14]

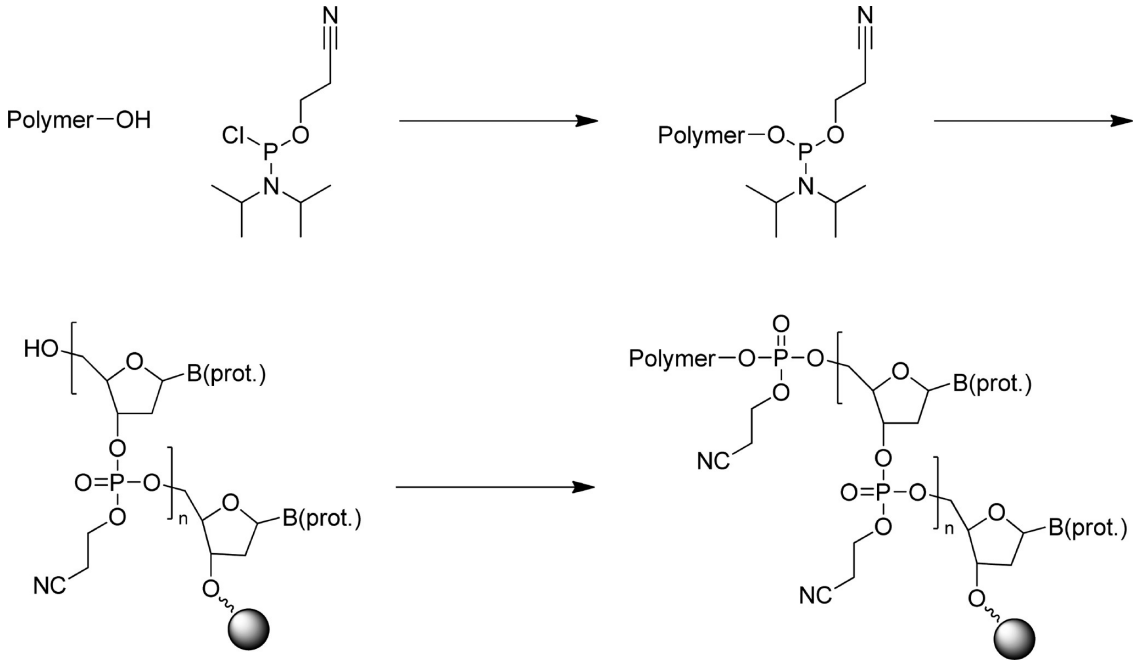
The appropriate selection of organic polymer and DNA sequence allows tuning and controlling of the material properties for designated applications. The introduction of hydrophobic polymers into DBCs leads, for example, to amphiphilic structures that self-assemble into micellar structures with hydrophobic core and hydrophilic DNA corona.[15] In the beginning, the synthesis of these molecular chimeras was based on chemical couplings in solution. However, this method gave access only to a limited set of DBC products since this synthetic route is limited to water-soluble polymers. Therefore, new and more sophisticated automated solid phase synthesis and molecular biology based methods were developed that overcame these restrictions and allowed the synthesis of amphiphilic DBCs. This Account will give an overview of the achievements made in our group regarding the synthesis and applications of DBCs in material science, biomedicine, and nanotechnology. Specific functions in these fields were enabled by their supramolecular self-assembly properties induced by microphase separation and Watson−Crick base-pairing, making these materials suitable for drug delivery, as scaffold for the assembly of biomaterials, or as addressable dispersion medium for carbon nanotubes.

## Synthesis of DNA Block Copolymers

Different grafting strategies allow the systematic variation of length and sequence composition of the nucleic acid segment as well as the nature and molecular weight of the polymer part. The coupling of prefabricated biological and chemical polymer building blocks through reaction of complementary terminal groups allows a straightforward synthesis of numerous linear DBCs. In general, two synthetic routes can be distinguished, coupling in solution and on solid support. Solution-based coupling reactions have up to now been limited to hydrophilic polymers since both reaction partners have to provide sufficient solubility in aqueous reaction media.

Different coupling reactions have been employed for the attachment in aqueous solution (Scheme 1). In the first

**SCHEME 2.** DBC Synthesis by Phosphoramadite Chemistry




approach, carboxylic acid terminated polymers were coupled to amino-functionalized ODNs forming an amide bond.[16,17]

A second strategy for solution-based conjugation of a hydrophilic polymer like poly(ethylene glycol) (PEG) to an oligonucleotide relied on disulfide bond formation between thiol-modified polymer and ODN.[18] Besides the formation of amide and disulfide bonds, the Michael addition was one of the common approaches for the attachment of ODNs to hydrophilic polymers. Acrylate- or maleimide-functionalized polymer moieties served as Michael acceptors, while thiol-modified ODNs acted as Michael donors.[19,20]

The copper-catalyzed Huisgen cyclo-addition, the most prominent member of the group of click-reactions, was recently applied for the coupling of DNA and polymer. The application of this pericyclic reaction in combination with DNA was limited by the fact that copper ions induce DNA strand breaks. With the introduction of a copper-chelating agent, the Huisgen reaction became an applicable and a straightforward method for DNA modification.[21]

All of these methods are well suited for the generation of DBCs with hydrophilic polymers. Since amino- and thiol-modified ODNs are available from commercial sources, the solution-based synthesis of DBCs is easily performed and can be carried out in a conventionally equipped chemical laboratory without the need for an expensive automated DNA synthesizer.

The yields of DBCs are drastically lower when hydrophobic polymers are coupled to ODNs by a solution-based process. The main reason for low coupling efficiencies is the different solubility properties of hydrophilic DNA and hydrophobic polymers in the reaction media.

For this reason, coupling strategies based on solid-phase synthesis approaches were successfully developed. First steps in this direction were performed by Mirkin and co-workers.[22] The amphiphilic DBCs were prepared through solid-phase synthesis on controlled pore glass beads (CPG) in a similar manner to conventional oligonucleotide synthesis. The key reagent in this approach was a polymer phosphoramadite (Scheme 2), which was obtained by treating alcohol-terminated polystyrene with chlorophosphoramadite. The coupling of polystyrene–phosphoramadite with the 5′-hydroxyl group of the oligonucleotide strand was carried out using the "syringe synthesis technique".[23] In this method, the cartridge containing the CPG with the synthesized ODN, still protected and attached to the solid support, is removed from the synthesizer. Subsequently, two syringes are attached to the inlet and outlet of the column. Finally, polymer–phosphoamadite solution is manually pumped through the cartridge several times to achieve ODN–polymer conjugate formation. Alternatively, cartridges with presynthesized ODNs can be purchased from commercial suppliers avoiding the investment costs for a DNA synthesizer. After deprotection and cleavage of the DBC from the solid support with aqueous ammonia, the desired DNA–polystyrene diblock structure becomes soluble in DMF and can be easily

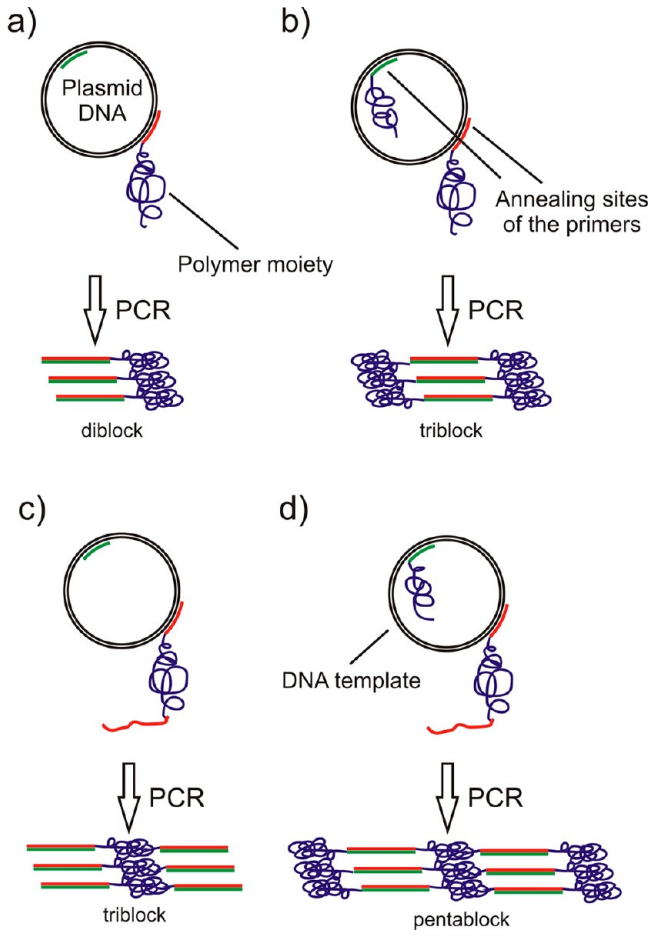



**FIGURE 1.** Schematic representation of the build-up of (a) DNA diblock, (b, c) DNA triblock, and (d) DNA pentablock copolymers by using PCR.

extracted from the CPG. This method employing syringes might have some drawbacks since high reproducibility and efficient exposure of the phosphoramidite polymer to the solid phase is not guaranteed.

To overcome these deficiencies, our group established the "in line" coupling of a polymer phosphoramadite with the detritylated 5′-hydroxyl end of a solid support bound ODN in a conventional DNA synthesizer.[24] The advantage over previous methods lies in the high reproducibility due to automation and the efficient exposure of the phosphoramadite polymer to the solid support. With this approach, it was, for example, possible to couple phosphoramadite–poly(propylene oxide) (PPO) derivatives to a 22-mer ODN.

The pure block copolymer of DNA-*b*-PPO was obtained after separation from the resin, deprotection, and purification by polyacrylamide gel electrophoresis (PAGE) or anion exchange chromatography. Throughout the text, we will denote block copolymers by naming the segments and separating them by a "*b*" indicating the *block*-type architecture. The coupling efficiencies of the large polymer moieties were remarkably high with yields reaching 41% and 32% for hydrophobic PPO polymers with molecular weights of 1000 and 6800 g/mol, respectively. This experimental outcome of the coupling procedure shows the superiority of the fully automated approach in comparison to a grafting approach in solution or a manual attachment procedure. The route of preparing DBCs fully "inline" gave also access to more complex DNA–polymer hybrid materials like triblock architectures of the type DNA-*b*-polymer-*b*-DNA.[25]

## Fabrication of DBCs by Molecular Biology Techniques

Besides chemical synthesis, molecular biology techniques became important tools for the generation and postsynthetic modification of DBCs. In the chemical routes, the length of the nucleic-acid segment of DBCs is restrained due to the limits of solid-phase ODN synthesis. These restrictions were successfully overcome by the application of molecular biology techniques like polymerase chain reaction (PCR) and enzymatic restriction and ligation.

PCR, one of the most versatile tools in molecular biology, was successfully applied for the preparation of DBCs with extended double-stranded (ds) nucleic acid blocks. For in vitro production of specific DNA sequences, a DNA template, two oligonucleotide primers, the four deoxynucleoside triphosphates (dNTPs), and a thermostable DNA polymerase are needed. In combination with single-stranded (ss) DBCs as primers, PCR has been successfully employed for the preparation of di-, tri- and pentablock architectures. The organic polymer blocks have been selected to be hydrophilic, hydrophobic, and thermoresponsive, proving the generality of PCR as a tool for the synthesis of well-defined block copolymer architectures with high molecular weights and monodisperse biological polymer units.[26,27]

By utilizing a ssDNA block copolymer and a conventional oligonucleotide as primers, diblock copolymers were obtained (Figure 1a), whereas the use of two ssDNA block copolymers as priming species resulted in triblock architectures (Figure 1b). While the latter approach yielded structures of type polymer-*b*-DNA-*b*-polymer, an architecture with a block arrangement of type DNA-*b*-PEG-*b*-DNA was realized by employing a sense primer ssDNA-*b*-polymer-*b*-ssDNA and an unmodified ODN antisense primer (Figure 1c). The triblock primer gave access to more complex block architectures. Pentablock structures, for example, of the type PEG-*b*-DNA-*b*-PEG-*b*-DNA-*b*-PEG are accessible by combination of two primers, a PEG-*b*-ssDNA and the before-mentioned triblock primer (Figure 1d). With this approach, we were able to

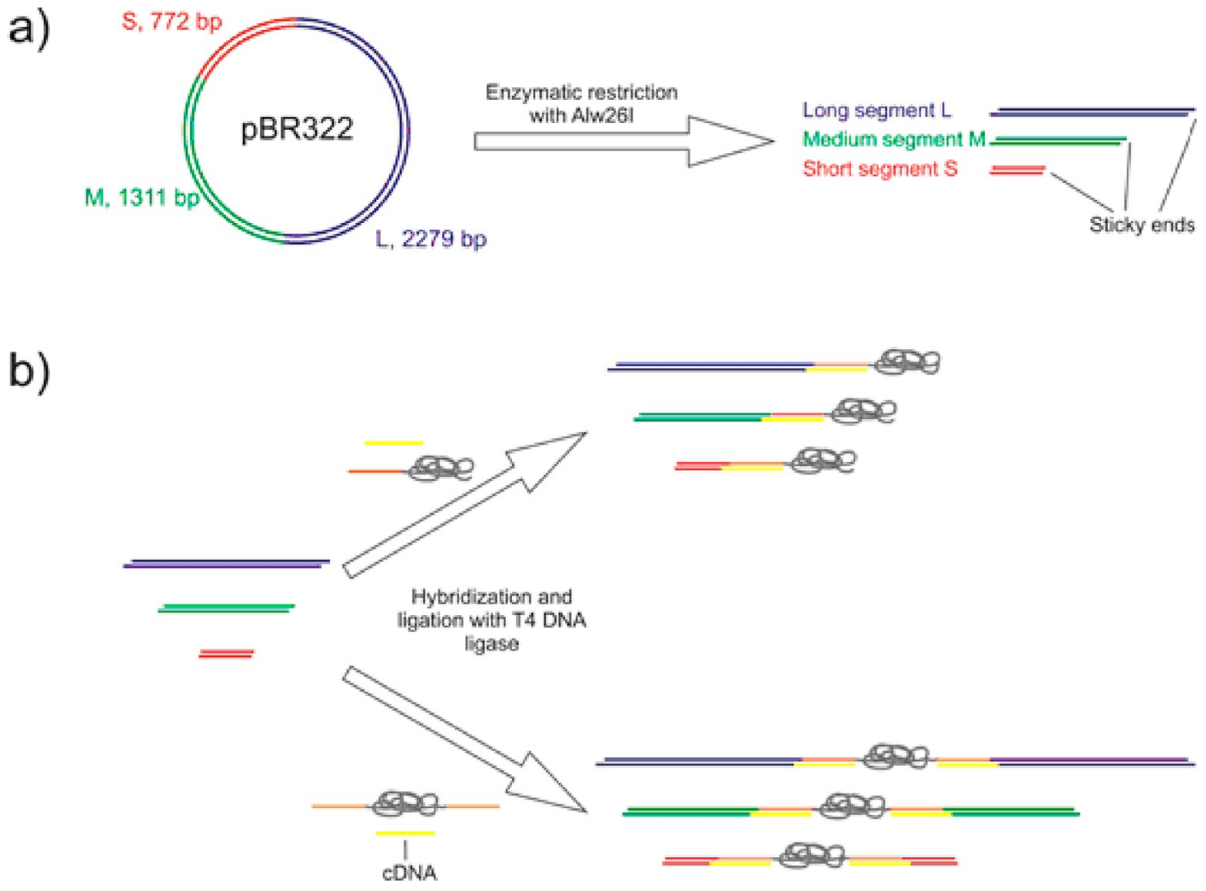



**FIGURE 2.** (a) Digestion of pBR322 by a DNA restriction enzyme, *Alw*26I, results in three dsDNA segments of different length. The four-nucleotide sticky ends are complementary to the 5′-overhangs of corresponding phosphorylated cDNA (yellow, shown in panel b). (b) Hybridization and ligation of ssDNA di- and triblock copolymers (DNA part orange, polymer moiety gray) with the appropriate cDNA and dsDNA segments in the presence of T4 DNA ligase results in extended di- and triblock copolymer architectures.

generate pentablock DBCs with monodisperse DNA segment lengths of 167, 225, and 500 base pairs that exhibit molecular weights up to 600 000 g/mol.[27] These results are remarkable from the perspective of a polymer chemist, since it was possible to generate ultrahigh molecular weight block copolymers with perfect structural composition.

Another approach based on molecular biology made use of restriction and ligation enzymes and allowed the generation of linear dsDNA di- and triblock copolymers that contain several thousand base pairs in the DNA segment.[28] For this purpose, ss di- and triblock DBCs were prepared by standard methods as described before. The diblock DBCs consisted of a 22mer ODN segment with the 5′-end attached to a polymer block. To show the generality of the approach regarding the organic polymer, we employed three different polymers of different length and polarity (PEG, PPO, and poly(*N*-isopropylacrylamide) (PNIPAM)). For the synthesis of triblock architectures, two identical ODNs were attached via their 5′ end to both ends of a PEG unit (Figure 2a). The long dsDNA parts for the extension of the di- and triblock DBCs were generated by digestion of circular plasmid DNA (pBR322) employing the DNA restriction enzyme *Alw*26I. As a result three dsDNA segments of different length were obtained (Figure 2b). Hybridization of ssDBCs with complementary sequences (cDNA) designed to introduce short overhangs and enzymatic ligation with the dsDNA restriction products were carried out in one pot through simple mixing and incubation resulting in the controlled extension of the DBCs (Figure 3c). With this technique, it was possible to generate nucleic acid extended DBCs with up to five times higher molecular weight (up to three million dalton) compared with the PCR approach. Since the sequence of the attached dsDNA strand is known, further manipulation of the structures is easily possible by digestion with other restriction endonucleases. This procedure enables alteration of high molecular weight DBCs with single nucleotide precision.

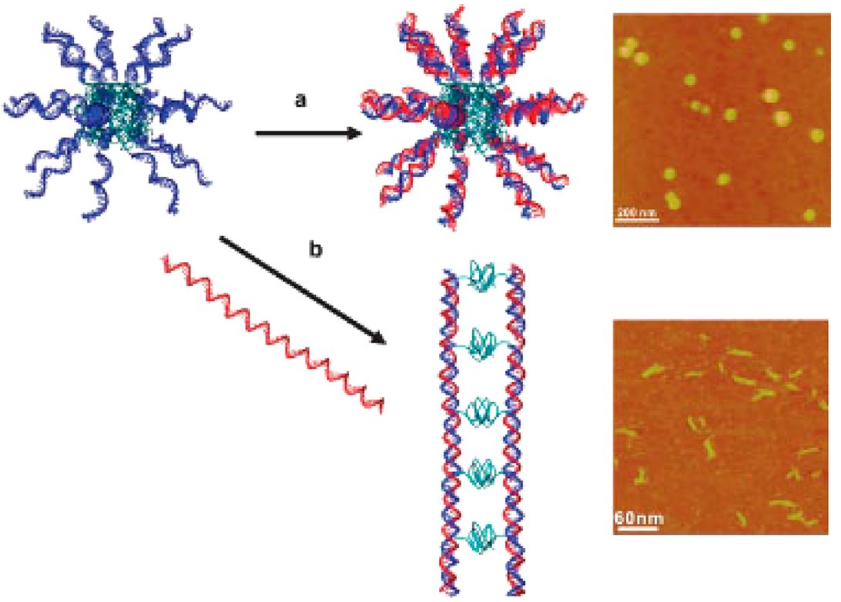



**FIGURE 3.** Schematic representation of hybridization of ssDNA-*b*-PPO micelles with different DNA molecules and corresponding AFM pictures. (a) Base pairing with a short complementary sequence yields micelles with a ds corona. (b) Hybridization with long DNA templates results in rod-like micelles consisting of two parallel arranged helices induced by hydrophobic interactions of the organic polymers (adapted from ref 34).

## Supramolecular Assemblies of DBCs

Amphiphilic block copolymers that consist of hydrophobic and hydrophilic parts are known to form various supramolecular structures like monolayers, micelles, or vesicles.[29−31] Such superstructures formed by polymer amphiphiles find application as containers for catalytically active species or as drug delivery vehicles. The use of DNA−polymer hybrid materials offers new opportunities compared with all organic block copolymers or peptide−polymer hybrides. Besides the automated and straightforward synthesis, the functionality of the synthetically programmable DNA part represents the key benefit of DBC materials. By introduction of the nucleic acid units, the amphiphiles possess addressable segments capable of Watson−Crick base pairing. This allows the formation of supramolecular nanostructures as well as convenient functionalization of the nanosized objects by hybridization with complementary ssDNA. Moreover, dynamic assemblies are accessible because the hybridization event itself can be exploited for morphological changes of DBC aggregates.

A simple example of such a supramolecular assembly is the formation of triblock copolymer structures of the type polymer-*b*-DNA-*b*-polymer from two DBCs with complementary sequences by duplex formation.[25] The same concept was extended to pentablock architectures by the assembly of a single-stranded triblock structure of the type DNA-*b*-PEG-*b*-DNA with two complementary diblock structures of type DNA-*b*-PEG. Large one-dimensional polymer assemblies were achieved when two ss self-complementary triblock structures with a DNA-*b*-polymer-*b*-DNA topology were employed.[32] The molecular weight of these supramolecular polymers could be easily controlled by the ratios of DNA−hybrid monomers since access of one monomer efficiently induced chain termination.

Besides extended one-dimensional assemblies, DBCs adopted micellar morphologies.[24] This is especially true for DBCs with hydrophobic polymer segments. In this case, the assembly of the DBCs was determined by the polymer moiety, which forms the hydrophobic core, while the DNA was present in the corona. When ssDNA-*b*-polystyrene (PS) micelles were fabricated, a cosolvent was needed for the micellization process,[22] while in case of DNA-*b*-PPO micelles, a simple heating and cooling protocol was sufficient due to the low glass transition temperature of PPO ($T_G = -70$ °C) compared with PS ($T_G = 95$ °C).

The structural properties of DBC micelles could be switched by hybridization. While base pairing of short ODNs to the corona did not change the spherical shape, hybridization with long templates induced the formation of rod-like aggregates (Figure 3).[33] Therefore, a template was designed that encodes the complementary sequence of the DBC micelle corona several times. During hybridization, a double helix was formed from which a PPO moiety protruded every 22 nucleotides. This led to the parallel arrangement of two double helices mediated by interactions of the hydrophobic polymer segments. The length of the rod-like aggregates was controlled by the size of the template.

Besides preparing supramolecular aggregates of pristine DBCs, it is possible to include other polymers as well. Pluronic block copolymers gained recently much attention as materials for biomedical applications.[35] The block copolymers are composed of PEG and PPO blocks arranged in a triblock architecture $PEG_n$-*b*-$PPO_m$-*b*-$PEG_n$. Due to their amphiphilic structure, Pluronic block copolymers form micellar structures similar to DBCs. The micelles can be cross-linked either at the periphery of the corona or within the core to protect them against disaggregation upon dilution and precipitation at low temperatures. While the PEG moieties within the micelle corona provide biocompatibility, the hydrophobic core allows loading of hydrophobic drugs. The primary limitation of Pluronic-based drug delivery system is the difficulty of functionalization, for example with targeting units. Such moieties can only be introduced by chemical modification of the PEG termini. DBC micelles on the other hand allow easy functionalization through simple hybridization with cDNA that is covalently linked to the desired functionality. *In vivo* applications of DBCs, however, might be limited by the instability of the micelles against dilution and the immune response, induced by the high local DNA concentration.

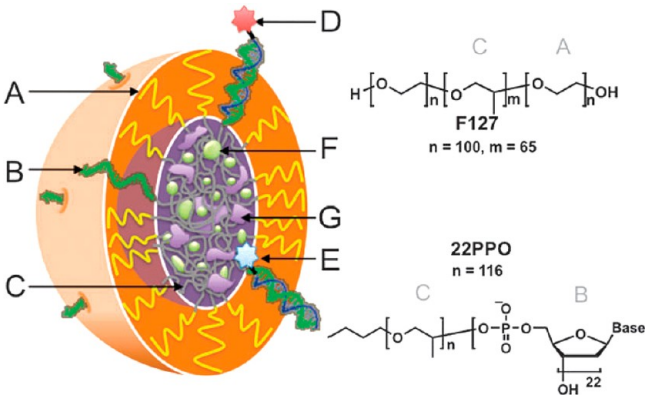



**FIGURE 4.** Schematic of the mixed micelle architecture and chemical structures of the polymeric components. (A) PEG block of Pluronic. (B) DNA block of DBC. (C) PPO blocks of Pluronic and DBC. (D, E) Probes at 5′- and 3′-ends of the cDNA, respectively. (F) Hydrophobic compound loaded into the hydrophobic core. (G) Cross-linked nanodomains of PETA in the core (adapted from ref 36).

By combination of Pluronic block copolymers and DBCs in mixed micelles, the shortcomings of both systems can be overcome. Mixed micelles of both block copolymers provide the sequence specific addressability of DBC, while the PEG chains might implement a kind of "stealth" function. To prove this concept, mixed micelles of DNA-*b*-PPO and Pluronic F127 were prepared by simply mixing both block copolymers (Figure 4). It was possible to stabilize these aggregates through UV-induced cross-linking of pentaerythrol tetraacrylate (PETA) in the core, as confirmed by fluorescence studies and AFM measurements. The mixed nature of the micelles was additionally proven by FRET studies between FAM–cDNA conjugates and TAMRA-labeled F127. Moreover, the controlled aggregation of gold nanoparticles through DNA hybridization confirmed the addressability and availability of the DNA in the corona for postsynthetic functionalization. The successful combination of both block copolymers within one micelle system paves the way toward further *in vivo* studies regarding drug release, targeted delivery, circulation lifetimes, and immunogenicity.

Besides micellar structures, vesicles with an average diameter of 80 nm and a closed double-layer membrane were formed by 12mer ODNs attached to polybutadiene.[37] These vesicles found application in adhesion and biofilm growth studies of *Escherichia coli*.[38]

## DBC Micelles as Drug Delivery Vehicles

While the mixed micelles are very promising candidates to be tested in drug delivery applications initial success in this area was achieved with pristine DBC materials. The selective

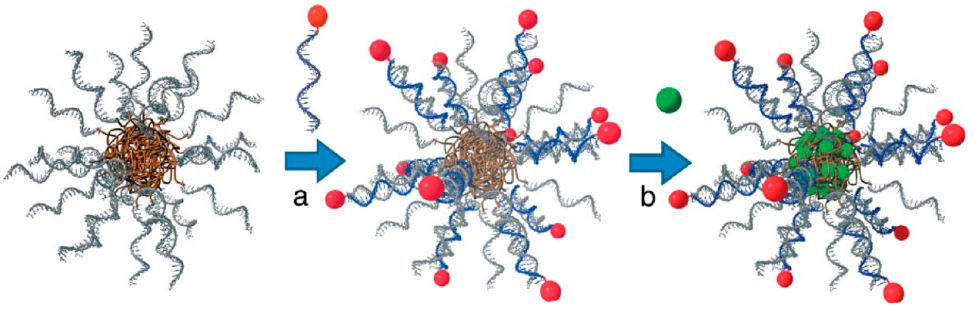



**FIGURE 5.** Schematic representation of the drug delivery system based on DNA block copolymers. (a) Targeting units (red dots) that are connected to the complementary sequence of the micelles are hybridized to equip the nanoparticle surface with FA units. (b) The anticancer drug (green dots) is loaded into the core of the micelles. Due to hydrophobic interactions of Dox with PPO, the drug accumulates in the interior of the DBC aggregates (adapted from ref 39).

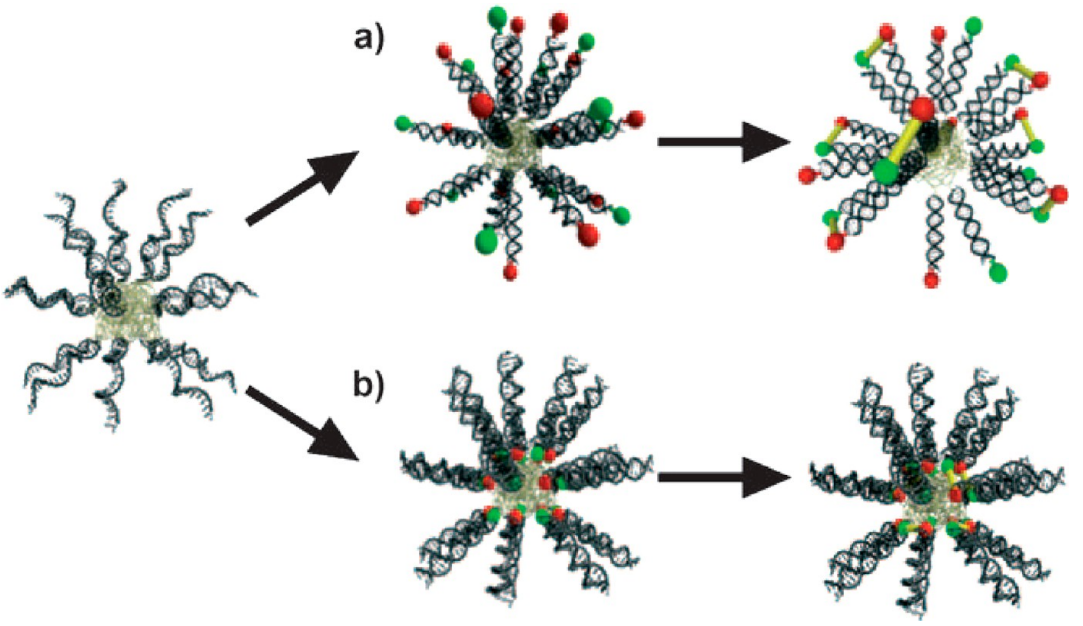



**FIGURE 6.** DBC micelles as scaffold for DNA-templated reactions. Single-stranded micelles are hybridized with oligonucleotides that are equipped with reactants either at the 5′- or at the 3′-ends. The subsequent chemical reaction (yellow bar represents a new bond) proceeds at the surface of the micelle (a) or at the hydrophobic/hydrophilic interface (b) (adapted from ref 24).

targeting of a specific organ or tissue is a major goal in drug delivery and bioimaging. Especially the specific targeting of cancer cells is of great importance since chemotherapeutic agents do not distinguish between cancerous and healthy cells, leading to systemic toxicity and side effects.

Amphiphilic DBCs forming micellar nanoparticles by microphase separation were successfully tested *in vitro* as drug delivery vehicles. In our approach, the DNA does not serve a biological function but enables the convenient and fast generation of multifunctional particles, while the hydrophobic core can be loaded with hydrophobic anticancer drugs. We equipped the nanoscale objects with targeting units that recognize overexpressed cell surface receptors of cancerous human colon adenocarcinoma (Caco-2) cells, that is, folate receptors. The corresponding ligand, folic acid, could be easily introduced to the surface of DBCs by hybridization after attaching it to cDNA. In a second step, the core was loaded with the hydrophobic anticancer drug, doxorubicin (Dox) (Figure 5). The combination of both units, folic acid at the micelle surface and Dox in the hydrophobic core, resulted in efficient cytotoxicity and high mortality among the cancerous cells. In contrast, when the folic acid was located at the interface of core and DNA corona, the micelles were no longer recognized by cancer cells. It was furthermore shown, that the receptor-mediated endocytotic uptake of DBC nanoparticles was most efficient with the maximum folic acid units placed at the rim of the micelles.

Since the drug loading and functionalization with various target moieties is extremely easy and straightforward, these micelles are qualified to be used as a combinatorial platform for testing and high-throughput screening of drug delivery vehicles.

## DBC Micelles as Templates for Chemical Reactions and Virus Capsids

Besides taking advantage of the easy functionalization procedure of DBC micelles for drug delivery, this feature was exploited to render DBC aggregates as programmable nanoreactors. DNA-templated chemistry takes advantage of the remarkable self-recognition properties of DNA.[40] It serves as scaffold that allows defined spatial arrangement of DNA-bound reaction partners. We successfully applied this concept to DBC-based micelles.[24] For this purpose, DNA-*b*-PPO aggregates were hybridized with equimolar amounts of ssODNs that were equipped with various chemical reactants. Due to the close proximity of the reactive groups in the micelle confinement, several organic reactions could be successfully carried out (e.g., isoindole formation, Michael addition, or amide bond formation). It was even possible to control whether the transformations take place at the surface or within the interior of the particles. This site specificity was achieved by the attachment of reactants at either the 5′- or 3′-ends of the ODNs, respectively (Figure 6).

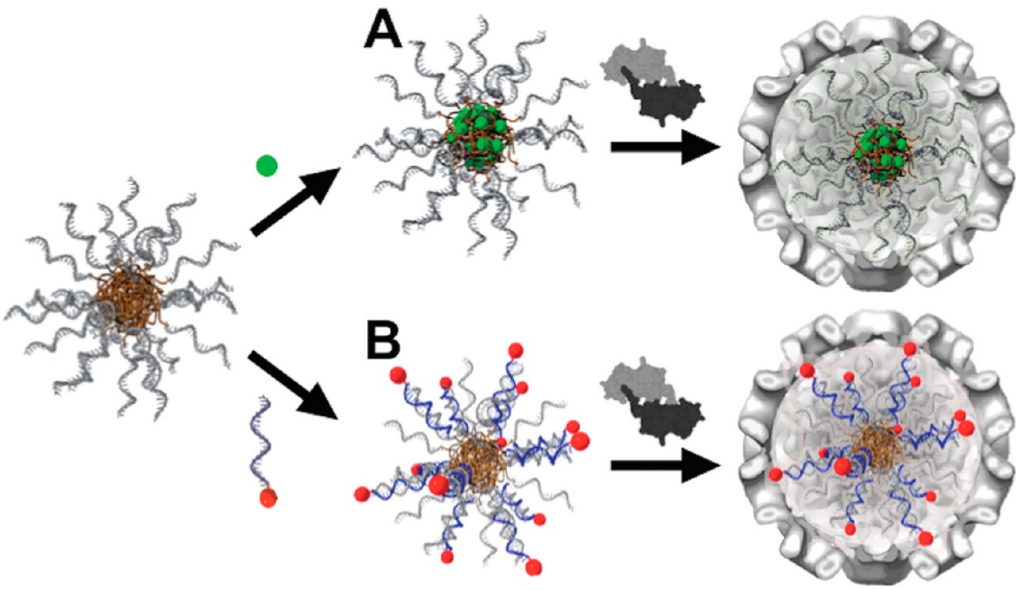



**FIGURE 7.** (a) Loading of hydrophobic molecules (green) into the core. (b) Equipping moieties (red) attached to cDNA by hybridization. Coat proteins encapsulate the micelle by a simple mixing process at neutral pH (adapted from ref 41).

For DNA-templated reactions, the specific interaction of micelle DNA with reactant DNA is crucial. In the following, it will be shown that the DNA in the DBC corona can also efficiently interact with proteins. In this regard, DBC micelles were employed as templates for virus capsid (VC) formation.[41] VCs or virus-like particles are a relative new class of natural biomaterials with great potential for materials science and nanotechnology. Especially their ability to specifically target individual cells has been attractive for gene delivery and has for quite some time now been harnessed for therapeutic delivery as well. The cowpea chlorotic mottle virus (CCMV) capsid is an excellent model system that is formed by the self-assembly of coat protein dimers. The loading of VCs is still a challenge since porous walls of the shell severely complicate loading and retention of small water-soluble molecules, while hydrophobic drugs are difficult to incorporate in these nanocontainers as well, due to solvent incompatibilities. The micellar aggregates of DBCs with their hydrophobic core and their anionic DNA corona overcome these shortcomings since they serve on the one hand as template for self-assembly of CCMV VCs and on the other as cargo carrier. Preloading of the micelles with hydrophobic entities in the core or hydrophilic entities by sequence specific hybridization enables the encapsulation of various small molecules inside VCs (Figure 7). The successful templating of CCMV VCs can be visualized by TEM micrographs of micelle-loaded VCs and bare DBC micelles (see Figure 8). With this general loading strategy, it might be possible to fully explore

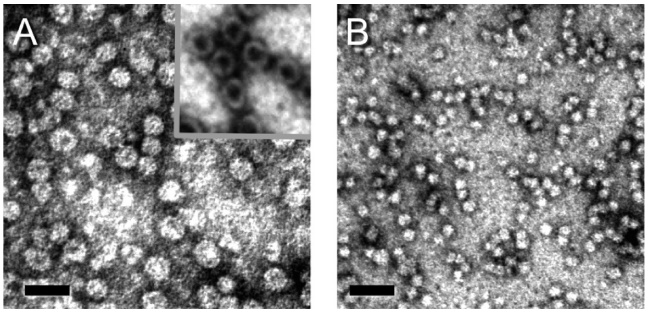



**FIGURE 8.** TEM micrographs, stained by uranyl acetate, of DBC-loaded VCs (A) and bare DBC micelles (B). The inset shows empty VCs for comparison obtained in a control experiment. Scale bars are 40 nm (adapted from ref 41).

diverse applications in view of VCs as high-impact drug delivery systems.

In another combination of different nanosized scaffolds, DBCs were employed with inorganic particles, in particular for the formation of DBC–gold nanoparticle networks. Therefore, DNA-modified Au nanoparticles were hybridized with DNA-*b*-PS micelles exhibiting the complementary sequence on the outer shell.[22] With this example, it was proven that DNA–polymer amphiphiles can be used to construct higher-ordered structures through sequence-specific hybridization with other nanomaterials that possess cDNA strands. Besides nanosized systems, the double-helix formation of DBCs was exploited for the preparation of microcapsules.[42] By alternating assembly of $T_{30}$-*b*-PNIPAM and $A_{30}$-*b*-PNIPAM layers around a silicon particle, subsequent PEG functionalization and removal of the silica core

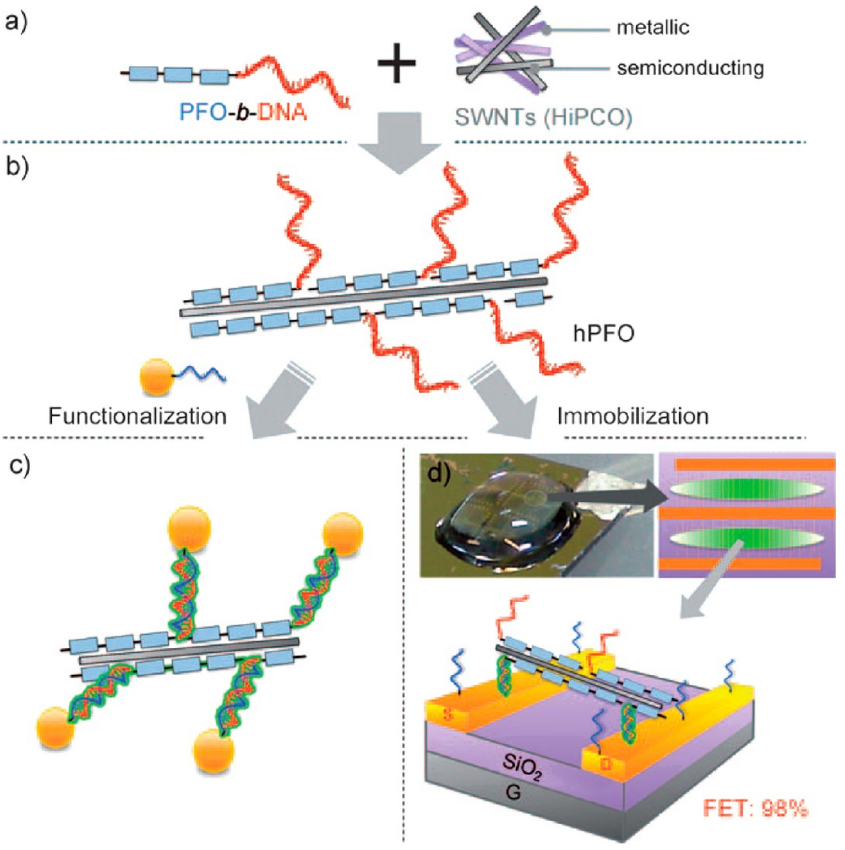



**FIGURE 9.** Dispersion and functionalization of SWNTs and their sequence-specific assembly by DBCs. (a) Amphiphilic DBC structure for solubilization of SWNTs. (b) Diameter-selective dispersion of SWNTs in aqueous media. (c) Straightforward functionalization of the SWNT surface by hybridization. (d) Unprecedented programmed assembly of SWNTs on surfaces or within nanoelectronic devices (adapted from ref 43).

particle, stable microcapsules were obtained. These hollow nanocontainers are envisioned to find applications in controlled delivery and release of therapeutics.

## Selection and Assembly of SWNTs

While most of the potential applications of DBCs are dedicated to the field of biomedicine, these materials might be equally important for nanoelectronics. Due to their outstanding mechanical and electronic properties, single-wall carbon nanotubes (SWNTs) play a key role in the development of modern functional materials. One of the major obstacles in handling and application of these promising new materials is their dispersion in solution and the isolation of SWNTs with narrow diameter window. The most easy and effective method for dispersion and separation is the application of dispersion agents. Poly(9,9-di-*n*-octylfluorenyl-2,7-diyl) (PFO) derivatives are even able to selectively solubilize certain semiconducting nanotube species in a narrow diameter range. Recently, nanotube field-effect transistors (FETs) gained much interest and are promising systems involving semiconducting SWNTs. The fabrication of such devices still involves the location and manipulation of individual SWNTs on a substrate, which is a complex and laborious task. Therefore, the development of a programmable, large scale positioning process for semiconducting carbon nanotubes was a major goal in device fabrication. An amphiphilic DBC consisting of a conjugated polymer and a ssDNA sequence acted as a dispersion agent for SWNTs and allowed solution of several of the key obstacles in SWNT technology, like nondestructive dispersion of individual nanotubes, enrichment of semiconducting tubes, and precise supramolecular addressability.[43] The DBC used for this approach consisted of PFO covalently bound to a 22mer ssODN. The hydrophobic polymer segment enabled the selective dispersion of semiconducting SWNTs, while the DNA block is still free for site-specific immobilization onto surfaces and facile hybridization with targets in solution. It was therefore possible to functionalize DBC-coated SWNTs with cDNA functionalized gold nanoparticles by sequence-specific Watson–Crick base pairing (Figure 9). The material also enabled the bottom-up fabrication of SWNT FETs in extremely high yields (98% working devices, Figure 9d).

Even without further processing and optimization, many of the produced transistors achieved a performance level comparable to reported individually fabricated SWNT-type devices with ambipolar conduction.

## Conclusion and Future Perspectives

The here presented applications and techniques show impressively the potential and versatility of DBC materials. A broad range of chemical and biological techniques can be used for the generation of monodisperse DNA blocks of virtually any length and sequence composition. In combination with the numerous coupling techniques for these molecules to hydrophilic and hydrophobic polymers, an infinite number of DBCs with a broad range of properties and characteristics becomes accessible.

The unique self-recognition properties of the DNA make DBC materials superior to other kind of block copolymers. In respect to self-assembled structures, hybridization with cDNA can be used for facile and specific functionalization; facilitating templated reactions, particle recognition, and the alignment of DNA-functionalized nanomaterials like SWNTs. Upcoming concepts from molecular biology like next generation methods for DNA assembly[44] or the design of adaptive materials by combination of DNA and organic polymers with specific DNA binding proteins[45] will have a distinct impact on the field of DBC materials. In combination with advanced analytical tools (like Förster related energy transfer) and visualization techniques (e.g., AFM, superresolution optical microscopy, and TEM), DBCs will give access to new functional materials and deliver insight into self-assembly processes of DNA nanomaterials.

The potential applications of these materials are located in such different areas as drug delivery, complex nanostructure fabrication, or nanoelectronics. Multidisciplinary approaches resulting from the interaction among researchers from different fields are indispensible to further evolution and realization of innovative concepts taking advantage of the unique properties of DNA block copolymers in high-tech applications.

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BIOGRAPHICAL INFORMATION

**Tobias Schnitzler** studied chemistry at the University of Mainz (Germany). From 2005 to 2009, he did his doctorate under supervision of Prof. K. Müllen at the Max Planck Institute for Polymer Research. In 2009, he joined the group of Prof. Herrmann as a postdoctoral fellow.

**Andreas Herrmann** currently holds a chair for Polymer Chemistry and Bioengineering at the Zernike Institute for Advanced Materials, University of Groningen, The Netherlands. He studied chemistry at the University of Mainz (Germany). In 2000, he completed his graduate studies at the Max Planck Institute for Polymer Research in Mainz. After a short stopover as a management consultant at Roland Berger, he returned to academia and worked as a postdoctoral researcher at the Swiss Federal Institute of Technology in Zurich on protein engineering. From 2004 to 2006, he was head of a junior research group at the Max Planck Institute for Polymer Research. In 2007, he received a call as full Professor at the University of Groningen.

FOOTNOTES

*To whom correspondence should be addressed. Mailing address: Zernike Institute for Advanced Materials, University of Gronigen, Nijenborgh 4, 9747 AG Groningen, The Netherlands. Telephone: +31(050)363 6318 4510. Fax: +31(050)363 6318 4400. E-mail: a.herrmann@rug.nl.

The authors declare no competing financial interest.

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