Selective DNA-Directed Assembly on Dual-Functionalized Microparticles

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Abstract—**The bottom-up assembly of functional devices requires novel building blocks to facilitate the incorporation of functional and structural hierarchy. Anisotropic building blocks can substantially broaden the creation of self-assembled devices with unique properties because of their morphological and/or chemical asymmetry. In this regard, we have created microspheres with one hemispherical face exposing silica and the other exposing gold. These microspheres were formed by the shadow deposition of gold onto silica microspheres. The two chemical surfaces allowed use of different surface reactions—silane chemistry for the silica side and thiol chemistry for the gold side—for immobilizing different oligonucleotide sequences on each of the two faces. These dual-functionalized microspheres were used in the selective orthogonal assembly of fluorophore-tagged target oligonucleotides. The DNA-directed assembly was confirmed by confocal microscopy of the microspheres. In essence, employing DNA as the linker molecule, these "Janus" particles can be assembled into various novel 1-D, 2-D, and 3-D structures, which are difficult to realize using symmetric building blocks.**

Index Terms—**DNA attachment, directed-assembly, microparticles, bifunctionality.**

I. INTRODUCTION

DNA as a tool for directed assembly has received considerable attention in the last few years. The sequence-specific base pairing of complementary DNA strands provides a unique strategy for programming the assembly of objects at the nanometer scale. Since the hybridization between two DNA strands is due to hydrogen bonding, the assembly strategy is easily reversible and achieves minimum energy states. The availability of commercial DNA synthesizers and a wide variety of synthetic precursors provides routine access to thousands of different selectable DNA sequences and thus offers easy scaling to high-throughput schemes. Most of the work in this field began after seminal contributions from Mirkin et al. [1] and Alivisatos et al. [2]. DNA as a linker molecule has been used to achieve a variety of self-assembled structures. Mirkin et al. [1] showed that Au nanoparticles change their optical properties as they are linked together into aggregates by DNA hybridization. They also showed the controlled binary assembly of nanoparticles of two different sizes into regular structures [3]. Niemeyer et al. [4-6] have used DNA hybridization to assemble other biomolecules like proteins, enzymes etc. Keating et al. [7] demonstrated that assembling nanoparticles using DNA hybridization helps to enhance detection of hybridization events using SPR. Smith et al. [8] have shown that DNA hybridization and subsequent modification of the DNA material using enzymes can be used for DNA-based computation. Since, DNA molecules by themselves do not have high electrical conductivity, Ben-Yoseph et al. [9] have chemically metallized DNA strands to achieve small wires with almost Ohmic-type electrical behavior. Recently, Mirkin et al. [10] and Niemeyer et al. [11] have demonstrated ultra-sensitive detection of proteins using DNA-derivatized nanoparticles.

As a rule in self-assembly processes, the more complex the building block, the higher the level of organization possible in the self-assembled structures [12]. To achieve higher-order structures by DNA-based assembly, there have been few attempts to use asymmetric building blocks. Mann et al. [13] have used gold nanorods to demonstrate DNA-driven assembly into anisotropic 3D-aggregates. Natan et al. [14] have used multifunctional nanorods as their starting material and achieved assembly using DNA hybridization. With a goal toward producing greater hierarchical complexity in such structures assembled using DNA hybridization, we targeted the generation of a bifunctional sphere as our asymmetric building block. To generate this structure, we use a simple shadow deposition technique to achieve a hemi-spherical coating of gold onto silica microspheres.

The technique for producing hemispherical gold coatings by evaporation was first used by Takei et al. [15]. They have also used these dual-functional particles to create an optical

sensor for avidin-biotin binding [16]. Many other methods have also been detailed in literature to achieve selective coating of a material on one side of a sphere. Kawaguchi et al. [17, 18] have used various techniques including Langmuir-Blodgett to selectively functionalize one-half of spherical particles. Fitzmaurice et al. [19] have used a filtration-based technique, which relies on shadow deposition in the liquid phase. Delville et al. [20] have achieved dual functionality using laser photochemical deposition. Muller et al. [21] have used tri-block copolymers to create bifunctional 'Janus' particles. Applications of such bifunctional structures have been demonstrated in making ultra-hydrophobic surfaces [22], systems for gene delivery [23], and other purposes. Bifunctional particles with hydrophobic and hydrophilic faces are noted to show surfactant-like behavior and can stabilize emulsions [24].

The main objective of this study was to create bifunctional silica particles using the shadow deposition of gold to produce an asymmetry. More importantly, our aim was to achieve selective orthogonal assembly of DNA molecules onto the different faces of these particles via DNA hybridization. We accomplished this goal by functionalizing the silica side and the gold side with two different DNA molecules. These particles were then used with fluorophore-tagged target DNA molecules to demonstrate the orthogonal assembly. The selective assembly of DNA molecules onto these surfaces is the first step in the process of employing DNA-directed assembly for generating more complex structures.

II. EXPERIMENTAL

A. Materials

Silica microparticles (30-50 μm) were obtained from Polysciences, Inc. (Warrington, PA). Octyl trichlorosilane (C_8 silane), dithiothreitol (DTT), and 6-mercapto-1-hexanol (MCH) were obtained from Aldrich (Milwaukee, WI). Trichloroacetyl (TCA) protected ω-hydroxyundecyl trichlorosilane (TCA silane) was custom synthesized in our laboratory. Gold pellets were supplied by J&J Materials, Inc. (Neptune City, NJ), and chromium-coated tungsten filaments were supplied by R. D. Mathis and Company (Long Beach, CA). Acetonitrile, methanol, ethanol, acetone, sodium chloride, sodium citrate, EDTA, tris-HCl, and potassium carbonate (K_2CO_3) were obtained from Mallinckrodt (Paris, KE). Phosphoramidites and reagents used for oligonucleotide synthesis, and empty TWIST™ columns were obtained from Glen Research (Sterling, MA). Piperidine (20%) in dimethyl formamide was obtained from Protein Technologies, Inc (Tucson, AZ). Deionized (resistivity = 18.2 MΩ) water was provided by a MilliQ deionization system (Millipore Corp., Bedford, MA). Target oligonucleotides 5'–CAC AGG TCG CAT-TAMRA–3' (cA) and 5'–Cy5- TCA GTT ATG GTA–3' (cB) modified with the fluorophores tetramethyl

Rhodamine (TAMRA) and Cy5 respectively were obtained from Sigma-Genosys (Woodlands, TX). Thiolated oligonucleotide 5'-SH-$(CH_2)_6$ - GCC TGA TAC CAT AAC TGA-3' (oligo B) and 5'-SH-$(CH_2)_6$- TCA GTT ATG GTA TCA GGC –3' (oligo B1) ware obtained from Integrated DNA Technologies, Inc. (Coralville, IA). Centri-Spin™ columns for gel filtration were obtained from Princeton Separations (Adelphia, NJ).

B. Functionalizing silica surface

The microparticles were cleaned in a 12.5 wt-% NaOH solution in a 3:4 H_2O:EtOH for 2 h. Subsequently they were washed 3-4 times with deionized water and dried. The silanizing solution was prepared by adding octyl trichlorosilane (C_8 silane) and trichloroacetyl (TCA) protected ω-hydroxyundecyl trichlorosilane (TCA silane) in a 20:80 volume proportion to anhydrous toluene (>99.5%) to yield 1 μL/mL (~mM) solution of the silanes in toluene. The particles were immersed in the silanizing solution for 4 h. After 4 h, the particles were washed once with toluene and twice with acetone and dried.

To expose hydroxyl groups on the particle surfaces prior to DNA synthesis, the TCA protecting groups were removed by immersing the particles in a 5 mM K_2CO_3 solution in 1:1 H_2O: methanol for 15 min. Stepwise synthesis of oligonucleotide 5'–ATG CGA CCT GTG CGA CCA–3' (oligo A) was performed on the particles using a Applied Biosystems ABI-392/394 automated DNA synthesizer. Cyanoethyl groups present on the phosphorus linkages were removed by immersing the slides in a 20-wt% piperidine in dimethyl formamide (DMF) solution for 4 h. Protecting groups on the nucleic bases were removed by immersing the particles in a 0.05 M K_2CO_3 solution in anhydrous methanol for 4 h. To functionalize the particles with sequence non-complementary to the TAMRA-labeled target, 5'–AGC CAA TTG GCC TTT T–3' (oligo A1) was synthesized on the particles.

C. Gold Coating

The silica particles functionalized with oligo A or A1 were spread as a monolayer on a Petri dish. Chromium (~50 Å) and gold (~300 Å) were sequentially evaporated at 1 Å/s onto the particles in a diffusion-pumped vacuum chamber operating at 2 x 10^{-6} torr. The particles were briefly sonicated to release them from the surface. The gold-coated particles were imaged in a Philips XL30 FEG-ESEM. Optical microscopy experiments were performed on a Zeiss Axioskop in the reflectance mode.

D. Functionalizing gold surface

The thiolated oligonucleotides (oligos B & B1) were obtained from the vendor in the disulfide form. The disulfide group was reduced in a 0.05 M DTT solution for 30 min. Excess DTT was removed using gel filtration on Centri-Spin™ columns. The oligonucleotide solution was dilute to yield a 5 μM oligonucleotide concentration. The particles were added to this oligonucleotide solution and

stirred for more than 16 h. Oligo B was used to create gold surfaces with complementarity to the Cy5-labeled target while oligo B1 was used to make the gold surface non-complementary to the Cy5-labeled target. After 16 h, the particles were washed, and then immersed in a 7.5 mM MCH solution for 1 h to cap sites on the gold surface not coated with the SH-DNA.

E. Hybridization and Confocal Microscopy

Three kinds of particles were formed: complementary - complementary (C-C), non-complementary - complementary (NC-C), and complementary - non-complementary (C-NC). For C-C particles, both the silica and gold sides have sequences complementary to the respective target DNA molecules. For NC-C particles, the silica side bears a sequence non-complementary (oligo A1) to the TAMRA-labeled target (olgio cA) while the gold-side has a sequence (oligo B) complementary to the Cy5-labeled target (oligo cB). For the C-NC particles, the silica side bears a sequence complementary (oligo A) to the TAMRA-labeled target (oligo cA) while the gold-side has a sequence (oligo B1) non-complementary to the Cy5-labeled target (oligo cB). The particles functionalized with DNA on both the silica and gold sides were contacted with a solution containing the complementary target oligonucleotides (cA and cB) at 0.5 µM concentration in 3x SSC (0.5 M NaCl, 0.05 M sodium citrate) for more than 16 h. The particles were then washed successively with 0.3 M NaCl in 1x TE buffer, 0.1 M NaCl in 1x TE buffer, and 0.3 M NaCl in 1x TE buffer.

Confocal microscopy of the particles was conducted on a Zeiss Axiovert 100M using two He-Ne lasers (544 nm and 633 nm) and 63x oil objective.

III. RESULTS AND DISCUSSION

The silica particles were first silanized to make the surfaces reactive towards phosphoramidite synthesis. We used a system of mixed monolayers, consisting of C_8 silane and TCA silane. Our previous studies have shown that these two silanes can be used to achieve DNA immobilization at high surface densities with minimal non-specific adsorption [25]. Based on our previous results, we selected a silane composition of 20 vol% TCA and 80 vol% C_8 silane to achieve a high density of surface DNA molecules. After the silanization, the TCA protecting group was removed using a deprotection protocol that has been optimized to minimize damage to the silane surface. The particles were then placed in a DNA synthesizer to conduct the stepwise synthesis of DNA on their surface using phosphoramidite chemistry. The use of an automated oligonucleotide synthesizer gives flexibility for changing the sequence synthesized on the surface with great ease.

After DNA synthesis on the silica particles, the particles were spread as a monolayer on a petri dish and were coated with gold in a thermal evaporation system. Care was taken to avoid the formation of particle multilayers on the surface,
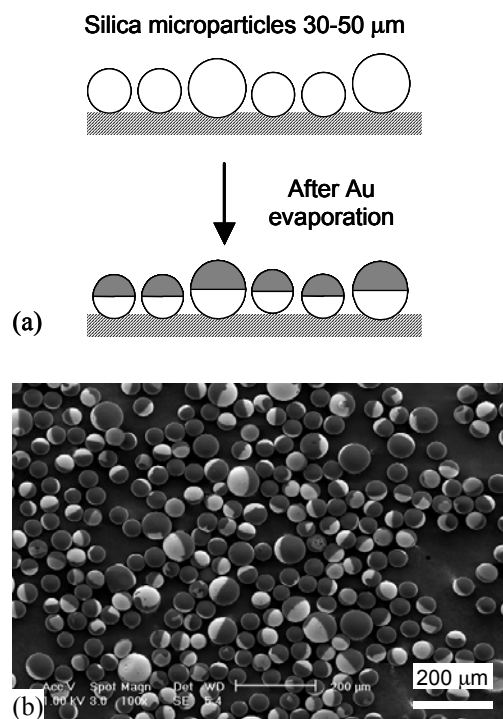

Fig. 1. (a) Schematic illustration of coating the silica particles (30-50 µm) with gold using shadow deposition. (b) SEM image of the dual-functional particles.

as they would reduce the efficiency of creating dual-functional spheres with perfect hemispherical coatings. Even after the gold deposition, the DNA synthesized on the uncoated half of the particles did not lose much of their activity as will be seen by the confocal microscopy results.

Fig. 1 shows a scheme for our shadow deposition of gold onto the silica particles and an SEM image of these coated particles in the backscatter mode. The regions coated with gold appear brighter because of the greater backscattering properties of the gold atoms. The image also shows that some particles do not have a hemispherical coating. This effect is due to formation of some particle multilayers that couldn't be avoided presently when spreading the particles on the flat surface before gold deposition. The protocol for this procedure could likely be optimized to reduce these defects in the future. Because of the differing wetting properties of the two surfaces, the dual-coated particles could be easily separated from the uncoated particles using a simple surface-pinning approach. Specifically, when the particles are placed on an air-water interface (Fig. 2a), before the particles are completely wet, most of the particles stay at the interface, as the interfacial tension is able to counteract gravitational forces. However, when the particles are stirred, the uncoated particles become completely wetted and settle to the bottom of the container (Fig. 2c) while most of the dual-functionalized particles remain at the interface (see Fig. 2b). The particles that stay at the interface also show an increased amount of clustering that occurs as a means to minimize their interfacial energy. This physical picture is further illustrated in Fig. 2d, where the
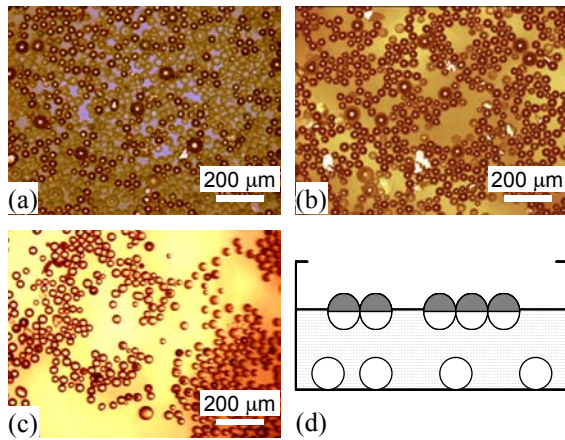
Fig. 2. Images of the dual functional particles taken under an optical microscope. (a) Image of the particles at the air-water interface after they are slowly sprinkled onto the water surface. Only the darker particles have a hemispherical gold coating. (b) Image of the particles at the air-water interface after gentle stirring. The particles remaining at the interface are mostly those with the hemispherical gold coating. (c) Image of flask bottom after gentle stirring. The uncoated particles settle to the bottom of the container under gravity. (d) Schematic illustration showing the pinning of the dual-functional particles at the air-water interface.

dual-functional particles become pinned at the air-water interface at the boundary of the gold coating. This behavior can be explained by the differing wetting properties of the two surfaces. The silica side is coated with negatively charged DNA molecules and thus is hydrophilic. The gold surface though inherently hydrophilic, readily picks up a layer of contamination because of its high surface energy and becomes hydrophobic. Wetting studies done on dual-functional particles by Veyssie et al. [26-28] also confirm the observed results. An important consequence is that we can quickly and easily separate the coated particles from the uncoated ones using this change in wettability.

The gold-coated particles were then treated with SH-DNA to functionalize the gold side selectively. The combination of the silica and gold surfaces ensures that the chemistries used for the surfaces have minimal amount of cross contamination by the other DNA sequence. Further, it is unlikely that the SH-DNA will affect the DNA on the silica side under the attachment conditions. Subsequently, the gold side was capped with MCH to reduce non-specific adsorption. Fig. 3 shows the overall scheme for making the dual-functional particles.

These particles were then immersed in a solution containing their fluorophore-tagged complementary target molecules. The target complementary to the silica side (oligo cA) was labeled with TAMRA and that to the gold side (oligo cB) was labeled with Cy5. The two fluorophores were selected to have minimal spectral overlap enabling the simultaneous detection of each fluorophore and allowing measurement of their respective spatial locations. To ensure the robustness of the assembly, we prepared three different kinds of particles: C-C, NC-C, and C-NC. C-C particles have sequences complementary (C) to the respective target DNA on both the silica and gold sides. The silica side for
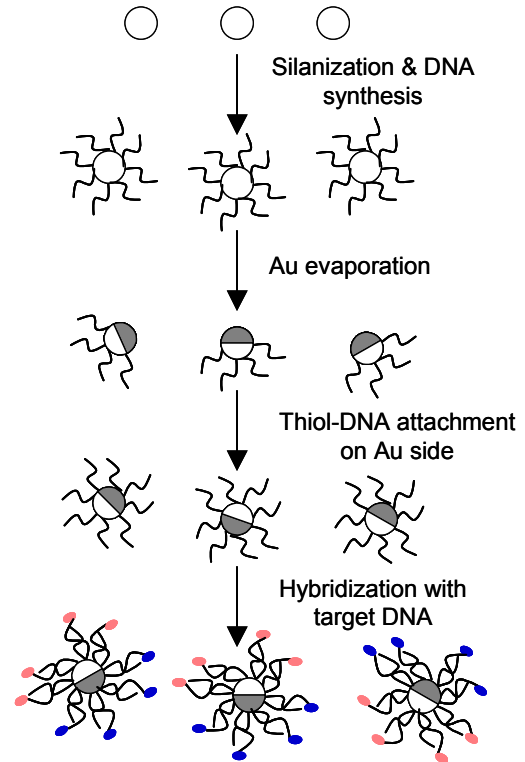

Fig. 3. Schematic illustration of the overall process of making the dual-functional particles and demonstrating the orthogonal assembly of fluorophore-tagged target DNA molecules on C-C particles. Briefly, the process begins with DNA immobilization on the silica particles, followed by Au deposition on one hemisphere, and SH-DNA attachment to the gold side. These dual-functional particles are then used for hybridizing with target molecules tagged with two different fluorophores (TAMRA & Cy5). The TAMRA-tagged target DNA is complementary to the DNA attached to the silica side and the Cy5-tagged target DNA is complementary to the DNA attached to the gold side.

NC-C particles and the gold side for the C-NC particles have sequences non-complementary (NC) to either of the target DNA molecules (oligo cA and cB).

Fig. 4 shows a representative fluorescence image of a cross-section of the three different kinds of particles as obtained by confocal microscopy. These 2-D images are of particles with exposed silica as the top-half and gold as the bottom half. For C-C particles (Fig. 4a), we observe a signal for both TAMRA and Cy5. For the C-NC particles (Fig. 4b), we observe that no fluorescence is observed on the Cy5 channel for both hemispheres, suggesting the efficiency of the DNA immobilization procedure for reducing non-specific adsorption; however, a signal is obtained on the TAMRA channel for the silica side indicating successful hybridization of the complementary sequences. For the NC-C particles (Fig. 4c), no Cy5 signal was obtained on the silica side but a very faint TAMRA signal was observed on the gold side, suggesting a small amount of non-specific adsorption on the gold side. These observations were corroborated for tens of particles of the three different types. Together, these confocal images show that we could achieve orthogonal assembly of two DNA molecules onto two specific regions determined by the base-pair
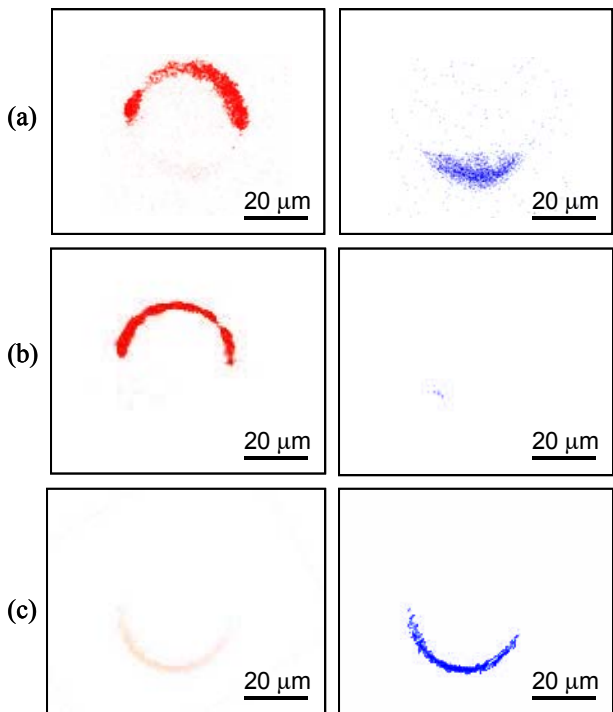
Fig. 4. Confocal microscopy images of cross-sections of dual-functional particles. The particles are oriented such that in these 2-D images the top half represents the silica side and the bottom half represents the gold side. The left column corresponds to the TAMRA signals and are obtained with a 543 nm laser excitation. The right column corresponds to the Cy5 signals and are obtained with a 633 nm laser excitation. TAMRA and Cy5 signals from   (a) a C-C particle with complementary sequences to the fluorophore-tagged target DNA molecules on both the silica side and the gold side. (b) a C-NC particle with complementary sequence on the silica side and non-complementary sequence on the gold side. (c) a NC-C particle with non-complementary sequence on the silica side and complementary sequence on the gold side.

complementarity of the DNA sequences. This study can be readily extended to achieve orthogonal assembly of nanoparticles by attaching the target oligonucleotides with nanoparticles. We are in the process of conducting the experiments to demonstrate the orthogonal assembly of nanoparticles and their assembly into highly complex hierarchical structures.

IV. CONCLUSIONS

Using a relatively simple system comprising of dual-functional microspheres generated by shadow evaporation of gold, we have demonstrated the orthogonal assembly of DNA molecules. We employed two different non-interfering chemistries to first achieve the orthogonal attachment of the DNA molecules onto the surfaces. These initial experiments on the orthogonal assembly of DNA molecules can be developed to achieve the self-assembly of complex hierarchical structures using DNA as the linker molecule between selectable particle-based building blocks.

REFERENCES

[1]   C. A. Mirkin, R. L. Letsinger, R. C. Mucic, and J. J. Storhoff, "A DNA-based method for rationally assembling nanoparticles into macroscopic materials," *Nature*, vol. 382, pp. 607-609, 1996.
[2]   A. P. Alivisatos, K. P. Johnsson, X. G. Peng, T. E. Wilson, C. J. Loweth, M. P. Bruchez, and P. G. Schultz, "Organization of 'nanocrystal molecules' using DNA," *Nature*, vol. 382, pp. 609-611, 1996.
[3]   R. C. Mucic, J. J. Storhoff, C. A. Mirkin, and R. L. Letsinger, "DNA-directed synthesis of binary nanoparticle network materials," *Journal of the American Chemical Society*, vol. 120, pp. 12674-12675, 1998.
[4]   C. M. Niemeyer, B. Ceyhan, and D. Blohm, "Functionalization of covalent DNA-streptavidin conjugates by means of biotinylated modulator components," *Bioconjugate Chemistry*, vol. 10, pp. 708-719, 1999.
[5]   C. M. Niemeyer, L. Boldt, B. Ceyhan, and D. Blohm, "DNA-directed immobilization: Efficient, reversible, and site-selective surface binding of proteins by means of covalent DNA-streptavidin conjugates," *Analytical Biochemistry*, vol. 268, pp. 54-63, 1999.
[6]   C. M. Niemeyer, "Progress in "engineering up" nanotechnology devices utilizing DNA as a construction material," *Applied Physics a-Materials Science & Processing*, vol. 68, pp. 119-124, 1999.
[7]   L. He, M. D. Musick, S. R. Nicewarner, F. G. Salinas, S. J. Benkovic, M. J. Natan, and C. D. Keating, "Colloidal Au-enhanced surface plasmon resonance for ultrasensitive detection of DNA hybridization," *Journal of the American Chemical Society*, vol. 122, pp. 9071-9077, 2000.
[8]   Q. H. Liu, L. M. Wang, A. G. Frutos, A. E. Condon, R. M. Corn, and L. M. Smith, "DNA computing on surfaces," *Nature*, vol. 403, pp. 175-179, 2000.
[9]   E. Braun, Y. Eichen, U. Sivan, and G. Ben-Yoseph, "DNA-templated assembly and electrode attachment of a conducting silver wire," *Nature*, vol. 391, pp. 775-778, 1998.
[10]   J. M. Nam, C. S. Thaxton, and C. A. Mirkin, "Nanoparticle-based bio-bar codes for the ultrasensitive detection of proteins," *Science*, vol. 301, pp. 1884-1886, 2003.
[11]   C. M. Niemeyer, R. Wacker, and M. Adler, "Combination of DNA-directed immobilization and immuno-PCR: very sensitive antigen detection by means of self-assembled DNA-protein conjugates," *Nucleic Acids Research*, vol. 31, 2003.
[12]   G. M. Whitesides and M. Boncheva, "Beyond molecules: Self-assembly of mesoscopic and macroscopic components," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 99, pp. 4769-4774, 2002.
[13]   E. Dujardin, L. B. Hsin, C. R. C. Wang, and S. Mann, "DNA-driven self-assembly of gold nanorods," *Chemical Communications*, pp. 1264-1265, 2001.
[14]   B. R. Martin, D. J. Dermody, B. D. Reiss, M. M. Fang, L. A. Lyon, M. J. Natan, and T. E. Mallouk, "Orthogonal self-assembly on colloidal gold-platinum nanorods," *Advanced Materials*, vol. 11, pp. 1021-1025, 1999.
[15]   H. Takei and N. Shimizu, "Gradient sensitive microscopic probes prepared by gold evaporation and chemisorption on latex spheres," *Langmuir*, vol. 13, pp. 1865-1868, 1997.
[16]   M. Himmelhaus and H. Takei, "Cap-shaped gold nanoparticles for an optical biosensor," *Sensors and Actuators B-Chemical*, vol. 63, pp. 24-30, 2000.
[17]   K. Nakahama, H. Kawaguchi, and K. Fujimoto, "A novel preparation of nonsymmetrical microspheres using the Langmuir-Blodgett technique," *Langmuir*, vol. 16, pp. 7882-7886, 2000.
[18]   K. Fujimoto, K. Nakahama, M. Shidara, and H. Kawaguchi, "Preparation of unsymmetrical microspheres at the interfaces," *Langmuir*, vol. 15, pp. 4630-4635, 1999.
[19]   L. Nagle, D. Ryan, S. Cobbe, and D. Fitzmaurice, "Templated nanoparticle assembly on the surface of a patterned nanosphere," *Nano Letters*, vol. 3, pp. 51-53, 2003.

[20] E. Hugonnot, A. Carles, M. H. Delville, P. Panizza, and J. P. Delville, ""Smart" surface dissymmetrization of microparticles driven by laser photochemical deposition," *Langmuir*, vol. 19, pp. 226-229, 2003.

[21] R. Erhardt, M. F. Zhang, A. Boker, H. Zettl, C. Abetz, P. Frederik, G. Krausch, V. Abetz, and A. H. E. Muller, "Amphiphilic Janus micelles with polystyrene and poly(methacrylic acid) hemispheres," *Journal of the American Chemical Society*, vol. 125, pp. 3260-3267, 2003.

[22] J. C. Love, B. D. Gates, D. B. Wolfe, K. E. Paul, and G. M. Whitesides, "Fabrication and wetting properties of metallic half-shells with submicron diameters," *Nano Letters*, vol. 2, pp. 891-894, 2002.

[23] A. K. Salem, P. C. Searson, and K. W. Leong, "Multifunctional nanorods for gene delivery," *Nature Materials*, vol. 2, pp. 668-671, 2003.

[24] B. P. Binsk and P. D. I. Fletcher, "Particles adsorbed at the oil-water interface: A theoretical comparison between spheres of uniform wettability and "Janus" particles," *Langmuir*, vol. 17, pp. 4708-4710, 2001.

[25] M. G. Bajaj, I. H. Lee, and P. E. Laibinis, in *preparation*.

[26] C. Casagrande and M. Veyssie, "Janus Beads - Realization and 1st Observation of Interfacial Properties," *Comptes Rendus De L Academie Des Sciences Serie Ii*, vol. 306, pp. 1423-1425, 1988.

[27] C. Casagrande, P. Fabre, E. Raphael, and M. Veyssie, "Janus Beads - Realization and Behavior At Water Oil Interfaces," *Europhysics Letters*, vol. 9, pp. 251-255, 1989.

[28] T. Ondarcuhu, P. Fabre, E. Raphael, and M. Veyssie, "Specific Properties of Amphiphilic Particles At Fluid Interfaces," *Journal De Physique*, vol. 51, pp. 1527-1536, 1990.