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Fabrication of Silica Nanomaterials Reflecting Morphological Transition of DNA Mediated by a Silane-appended Ionic Liquid

Kojiro Shimojo,* Hisayoshi Mitamura, Tsuyoshi Mouri, and Hirochika Naganawa
Division of Environment and Radiation Sciences, Nuclear Science and Engineering Directorate, Japan Atomic Energy Agency, Tokai-mura, Naka-gun, Ibaraki 319-1195

(Received January 21, 2011; CL-110052; E-mail: shimojo.kojiro@jaea.go.jp)

A simple fabrication strategy of DNA-transcribed silica materials has been developed. An ionic liquid incorporating an alkoxysilane group is capable of binding with DNA and acts as nuclei for subsequent sol–gel reaction with tetramethoxysilane. Unique silica materials with worm-like, rod-shaped, toroidal, or linearly fibrous structures are created from the same DNA template through the morphological transition of DNA induced by the ionic liquid.

The use of biological molecules and assemblies as templates has attracted much attention as biomimetic strategies for producing nanosized inorganic superstructures. Biological templates such as chitin,[1] collagen,[2] virusus,[3] and bacteria[4] have been utilized to direct the deposition of silica clusters, which enables transcription of sophisticated biological structures into silica materials. DNA is one of the most promising biomolecular templates because it has a refined double-helical structure that cannot be prepared in an artificial system. Furthermore, DNA condensation with multivalent cations allows morphological transition of DNA from the natural worm-like coil form to compact forms such as rod-shaped, toroidal, or spherical structures.[5] If DNA could be used as a template for sol–gel reactions under various conditions, several unique silica structures reflecting morphological changes of DNA could be fabricated from the same DNA. However, the inactive binding ability between polyanionic DNA and negatively charged silica species makes it impossible for DNA to act as a template in general sol–gel reactions. To overcome that problem, Numata et al.[6] transformed DNA into an overall positively charged species through complexation with a cationic amphiphile bearing one ammonium group and one guanidinium group, which permitted deposition of anionic silica on the surface of the DNA–amphiphile complex via electrostatic interaction. In addition, Jin et al.[7] reported the directing synthesis of a DNA–silica complex by using the cooperative effect of *N*-trimethoxysilylpropyl-*N*,*N*,*N*-trimethylammonium chloride. Reports on fabrication of silica superstructures using DNA template are fairly rare, and more general strategies are needed to expand the versatility of DNA transcription into silica materials. Herein, we demonstrate a new one-pot synthesis of DNA-transcribed silica materials utilizing the interaction between DNA and an ionic liquid (IL) incorporating an alkoxysilane group.

ILs, which have chemical and physical properties that are different from those of molecular liquids, have received growing attention as a new class of solvents in many fields of chemistry and industry. The unique properties of ILs offer great potential as reaction media in separation science[8] and biological chemistry.[9] Recently, several research groups reported studies on the binding between typical alkylmethylimidazolium ILs and DNA:[10] the imidazolium cations of ILs interact with the phosphate groups of DNA through electrostatic attraction, and the alkyl chains of cationic groups interact with the bases through strong hydrophobic association. Introduction of functional groups into ILs (task-specific ILs[11]) gives rise to marked changes of their properties, allowing a wide variety of applications beyond their use as solvents. In the present study, we synthesized a task-specific IL incorporating an alkoxysilane moiety[12]: (1-methyl-3-(3-trimethoxysilylpropyl)imidazolium chloride, [TMOSmim][Cl], synthetic procedure in Supporting Information (SI)[13]). In sol–gel reactions using DNA as a template, this IL is expected to play key roles: functioning as a mediator for the interaction with negatively charged DNA, as nuclei in sol–gel reactions, and as a condensing agent, inducing morphological transition of DNA from the natural worm-like coil form to rod-shaped and toroidal structures. In this study, we aimed to fabricate DNA-transcribed silica superstructures by means of the one-pot synthesis shown in Figure 1. By binding of $TMOSmim^+$ into DNA, the alkoxysilane sites are arranged on the surface of DNA, and subsequent polycondensation with tetramethoxysilane (TMOS) enables transcription of DNA into silica materials with worm-like, rod-shaped, toroidal, or linearly fibrous structures. Detailed experimental procedure for the sol–gel reaction is described in the SI.[13] The DNA transcription into silica was carried out under conditions such that the [TMOSmim][Cl] concentration was kept at 10 mM. The stock solutions of [TMOSmim][Cl] and TMOS were prepared from methanol to prevent the sol–gel reaction from occurring spontaneously.

In a preliminary experiment, we measured the UV and CD spectra of λ-DNA (48502 bp) at different pHs (Figure S1, SI[13]) and confirmed that morphological transition of λ-DNA was not caused in the pH range from 5.5 to 11.0 without [TMOSmim][Cl] and TMOS. Figure 2 shows TEM images of

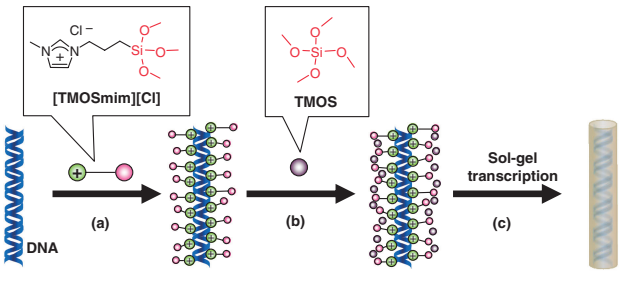

Figure 1. Schematic illustration of DNA transcription into silica. (a) Attachment of silane sites to DNA surface through binding with [TMOSmim][Cl], (b) addition of TMOS, (c) synthesis of DNA-transcribed silica materials by sol–gel reaction.

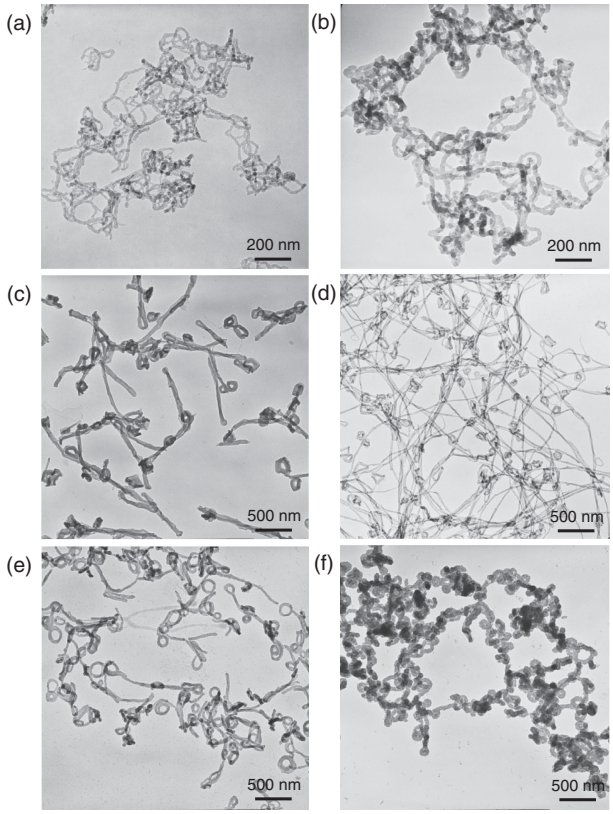

Figure 2. TEM images of silica materials with worm-like, rod-shaped, toroidal, or linearly-fibrous structures created by using λ-DNA template at different pHs and TMOS concentrations. (a) pH 8.0, [TMOS] = 11 mM; (b) pH 8.0, [TMOS] = 22 mM; (c) pH 5.5, [TMOS] = 11 mM; (d) pH 5.5, [TMOS] = 22 mM; (e) pH 11.0, [TMOS] = 11 mM; (f) pH 11.0, [TMOS] = 22 mM. [λ-DNA] = 57 ng μL^{-1}, [TMOSmim][Cl] concentration = 10 mM.

silica materials fabricated by using λ-DNA template at different pHs and TMOS concentrations (see SI,[13] Figure S2, for close-up images with same scale). As shown in Figure 2a, the sol–gel reaction in a neutral solution (pH 8.0) with 11 mM TMOS generated worm-like fibers 12–18 nm wide, which tended to knot with themselves or with adjacent fibers. The worm-like fibers originate from the random structure of λ-DNA. Furthermore, energy-dispersive X-ray (EDX) spectroscopy elemental mapping and point analyses confirmed that the worm-like fibers were composed of silica, characterized by the distribution of C, O, and Si atoms (Figure S3, SI[13]). The results convince us that $TMOSmim^+$ cations arranged on the surface of λ-DNA act as nuclei for the sol–gel reaction and that subsequent polycondensation with TMOS allows transcription of λ-DNA morphologies into silica. When the TMOS concentration was increased to 22 mM, the width of the silica fibers increased to 23–28 nm by further growth (Figure 2b). Additionally, removal of DNA template from silica materials by calcination was carried out. To obtain robust silica materials and enhance the image contrast between silica and hollow, the sol–gel reaction was conducted with the TMOS concentration at 33 mM. Hollow silica tubes (i.d. $\approx$4 nm) were partially observed by TEM, which is consistent with the width of λ-DNA–$TMOSmim^+$ complex (Figure S4, SI[13]). This supports that DNA acts successfully as a template. Furthermore, the control tests concerning the binary systems DNA + TMOS, DNA + [TMOSmim][Cl], or [TMOSmim][Cl] + TMOS were carried out. Instead of DNA-transcribed silica materials, silica nanoparticles and their aggregates were observed (Figure S5, SI[13]).

In weakly acidic conditions (pH 5.5) with 11 mM TMOS, a nearly equimolar silica mixture of rod-shaped and toroidal structures was obtained (Figure 2c). The rods were 47–67 nm in width, and the toroids had similar thickness and holes with inner diameter of 21–43 nm. This indicates that $TMOSmim^+$ can efficiently stabilize the sharp bends and packing of linear DNA as a condensing agent, inducing the morphological transition of λ-DNA from the natural worm-like coil form to compact forms such as rod-shaped and toroidal structures. Interestingly, when the TMOS concentration was increased to 22 mM, the linearly fibrous structures were obtained instead of the rod-shaped structures. Also, the network formation by crossovers of the fibers and decrease in the width of silica structures (20–32 nm) were observed (Figure 2d). Generally, it is known that acid catalysts accelerate one-dimensional polycondensation with TMOS in sol–gel reactions.[14] It is assumed that increase of TMOS concentration at pH 5.5 can enhance one-dimensional growth of silica, inhibiting DNA compaction to the rod-shaped structures by $TMOSmim^+$. As a result, fibrous structures expanded linearly were produced.

When a basic solution (pH 11.0) with 11 mM TMOS was employed, a mixture of rod-shaped and toroidal silica compounds was formed (Figure 2e), with different structures from those obtained under the weakly acidic conditions. The rods formed under alkaline conditions had narrow width (26–49 nm) and short length, whereas the toroids were thin (22–54 nm thickness) and had large holes with inner diameter of 37–136 nm. The population of toroids was higher than that of rods. However, increasing the TMOS concentration resulted in the aggregation of thick fibers (46–61 nm) due to three-dimensional growth of silica by base-catalyzed polycondensation (Figure 2f).[14]

As described above, pH has a great influence on the structures of silica compounds. The degree of ionization of the phosphate groups in DNA increases with increasing pH (See SI,[13] Figure S6, for the agarose gel electrophoresis of λ-DNA at different pHs). Thus, the degree of binding of $TMOSmim^+$ with DNA would vary markedly with pH. Obviously, the extent of DNA compaction depends on the balance between the degree of ionization of DNA and the degree of binding of $TMOSmim^+$, which is an important factor dominating the structures of silica compounds. Considering the fact that multivalent cations can induce efficiently DNA condensation,[5] polycationization of $TMOSmim^+$ through sol–gel reaction by itself should be also related to the morphological transition of DNA. In addition, the growth process and reaction rate of sol–gel polycondensation, which are strongly dependent on pH, are likely to be other important factors in the morphological transition of DNA during the transcription process into silica gel.

In summary, we succeeded in fabricating unique silica nanomaterials that transcribed higher-order DNA superstructures through sol–gel reaction. The task-specific IL $TMOSmim^+$ cations arranged on the surface of DNA act as nuclei for subsequent sol–gel reaction with TMOS, which can mediate the transcription of DNA structures into silica materials. The various

silica compounds with worm-like, rod-shaped, toroidal, or linearly fibrous structures were created from the same DNA template through the morphological transition of DNA induced by $TMOSmim^{+}$. These findings will expand the frontiers in formation of more complicated, but sophisticated inorganic materials. Fabrication of silica nanosheet using two-dimensional DNA array templates formed by self-assembly of artificial oligonucleotides is currently under investigation.

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