Metal Cluster Deposition on Genetically Engineered Tobacco Mosaic Virus Biotemplates

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Functionalized nanomaterial synthesis from the nanoscaled biotemplates is a promising subject in microelectronics and bioengineering research for its potential applications [1]. Specifically, the hybrid material of discrete inorganic nanoparticles and biomolecules has widely been studied to materialize physical properties such as optical and electrical properties while maintaining the original properties of the composing inorganic nanoparticles. Biotemplate technique, where biomolecules are used as a template, provides a simple and effective way to control nanoparticles in a designed manner using biological reactivity [2]. As well as the reactivity, the natural complex structure of biomoleucles in a wide variety of sizes made the biomolecules proper substances for the nanoscaled templates. Recent progress in genetic engineering has made biomolecules more promising elements for the nanomaterial synthesis [3]. Using current biological technologies, several biomolecular strands have been used in nanowire and nanorod synthesis [4], and proteins and viruses in the crystalline phase were used in the fabrication of nanoparticle array and liquid crystal assemblies [5,6]. In this research, the conductive nanotube synthesis through the metal nanoparticle deposition onto genetically engineered tobacco mosaic virus (TMV) template is studied. The structure and surface property of wild type TMV were modified to obtain favorable properties for the metal nanoparticle deposition. Nanotubes were synthesized by the deposition of various metal nanoparticles onto these engineered virus particles and the synthesized nanotubes were applied to the electrical devices as conductors and tested their electrical conductivity.

Tobacco mosaic virus is a tubular plant virus with 300nm length and 18nm diameter. Due to its cylindrical structure and abundant carboxylates on the inner and outer surface, the wild-type TMV was used as a template for inorganic nanotube synthesis [7]. In the previous studies the charge-charge interaction between the negative TMV surface and metal cations was the driving force for metal particle binding on the surface [8]. Several studies showed synthesized nanotubes and nanorods composed of silicate oxide, lead sulfides, and other metal particles using the wild-type TMV template. However, the lack of reliable binding site for metal particles resulted in irregular and discrete coatings on the wild-type TMV, which made it hard to fabricate the coated TMV particles further. Also the 300nm length of the wild-type TMV was an obstacle in preparing longer nanotubes. To overcome these problems, the capsid proteins of TMV were genetically engineered via site-directed mutagenesis in this research.

Two different types of engineered TMV particles were prepared as templates; one is the surface-modified TMV, 2cysTMV, whose surface were modified with sulfhydryl groups. The sulfhydryl groups were introduced by the addition of two extra cysteines at N-terminus of capsid protein. The other one is a shape-controlled TMV, E50Q, whose average length is extended to ~900nm. In preparing these engineered TMV particles, the cDNA of TMV capsid protein was modified via mutagenesis and infectious RNA transcripts containing engineered

capsid protein were prepared. The leaves of Nicotiana tobaccom cv. Xanthi were inoculated with the RNA and the engineered TMV particles were harvested. The ribbon structure of the engineered TMV capsid proteins are shown in figure 1. For the surface-modified TMV, the extra cysteines are exposed to the outer surface providing strong binding site for the metal particles (figure 1a). The structure of the shape-controlled TMV capsid protein is given in figure 1b. The substituted amino acid resulted in elongated structure by reducing the electrostatic repulsion between neighboring capsid proteins in axial direction [9].

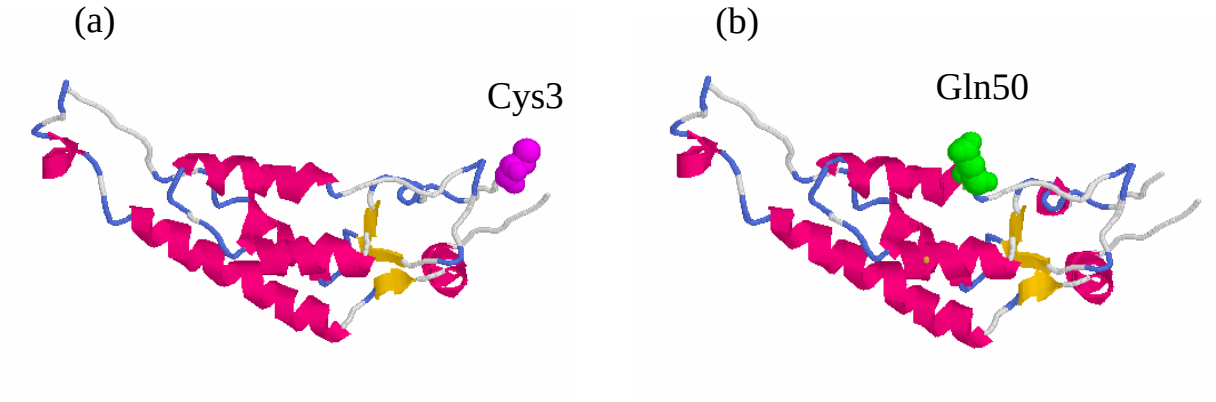

Figure 1. Ribbon structure of the engineered tobacco mosaic virus capsid proteins (a: 2cysTMV. Active sulfhydryl residues were exposed to the outer surface. b: E50Q. By substitution Glu with Gln, electrostatic repulsion between neighboring capsid proteins was reduced)

Metal nanoparticle deposition on the engineered TMV template was carried out by the metal salt reduction. Various metal salts were used as metal sources for the deposition. To the mixture of engineered TMV template and desired metal salt solution, reducing agent were added to form solid nanoparticles. After the addition of reducing agent, aliquots of the mixture were taken and positioned on the copper grid for investigation. The shapes of the synthesized metallic nanotubes and metal particle-decorated TMV particles were investigated using transmission electron microscopy (TEM), and chemical compositions were confirmed using electron dispersive spectroscopy (EDS). The electrical conductivity of the synthesized metallic nanotubes was measured by measuring currents with the swipe of applied voltage. The metallic nanotubes were positioned between two gold electrodes with ~50nm gap which was prepared by step junction technique [10]. The image of the bridging nanowire was taken with field emission scanning electron microscopy (FE-SEM).

Dense and smooth coating of metal nanoparticles was achieved using 2cysTMV template. Several metal particles like gold, silver, platinum, and palladium nanoparticles were deposited in a defined fashion onto the 2cysTMV surface. Comparing to the previous reports, it is clear that this improvement is due to the cysteine residues on the surface. The strong metal particle binding was confirmed from the binding metal particles after following washing and centrifugation procedures. When the wild-type TMV was used as a template, all the metal particles were washed off because of its weak binding force. The average metal particle size bound to the surface was also decreased when the 2cysTMV was used. The tiny particle formation on the 2cysTMV is thought to be due to the metal-thiolate complex formed in the

aqueous metal salt and TMV mixture [11]. The stable metal-thiolate complex worked as a nucleation site for the metal nanoparticle formation on the 2cysTMV surface. On the while, E50Q did not show any improvement in particle deposition. The same surface property of E50Q with that of wild-type TMV resulted in discrete metal particle deposition. However, the elongated TMV template provided a possibility for a micron-scale nanotube synthesis. TEM images of the platinum nanoparticle-deposited 2cysTMV and E50Q are given in figure 2. For the future application of the synthesized TMV nanotube, the electrical conductivity of metal-deposited nanotube was tested. Platinum-deposited 2cysTMV showed conducting behavior. This result provides a promising example of nanowire synthesized from the genetically engineered biotemplates. The I-V curve of the platinum-deposited 2cysTMV nanowire is given in figure 3.

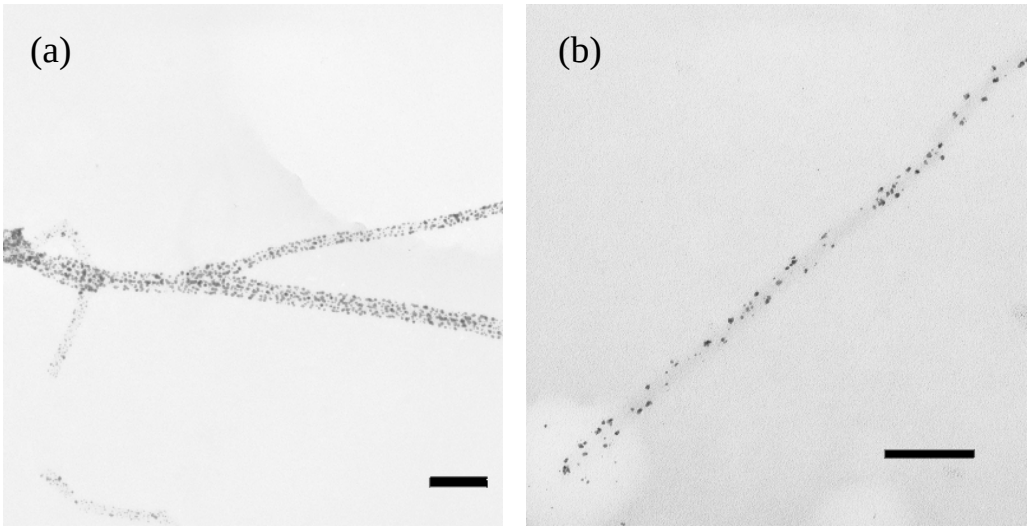

Figure 2. Platinum nanoparticle-deposited engineered TMV
(a: Pt-deposited 2cysTMV. b: Pt-deposited E50Q. Scale bar: 100nm)

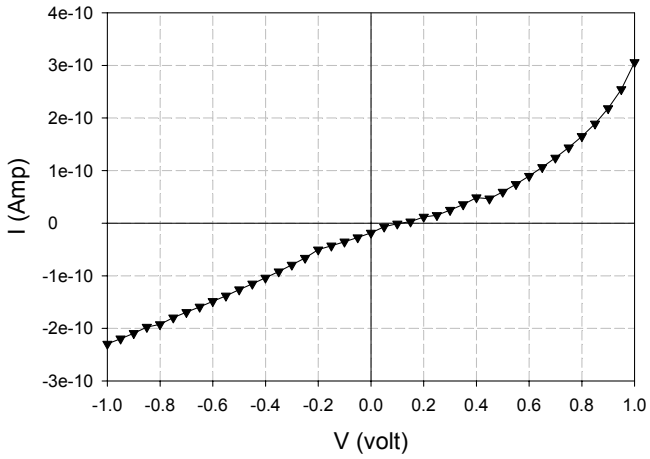

Figure 3. I-V curve of platinum-deposited 2cysTMV

In conclusion, improved deposition of various metal nanoparticles to the genetically engineered TMV template was demonstrated in this research. Extra cysteine residues provided strong binding site for the metal nanoparticles and resulted in well-defined metal particle coatings on the TMV templates. The metal coated TMV nanotubes were tested as conductive nanowires for the future application to the electrical devices. These results provides examples of surface functionality-designed (or shape-designed) biotemplates for the nanomaterial synthesis via genetic engineering combined with nanoparticle technology offering a better way for the metallic nanowire and nanotube synthesis.

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