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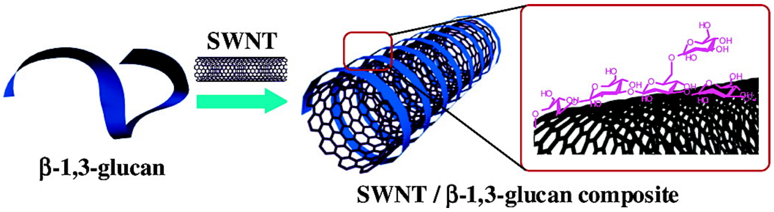

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Inclusion of Cut and As-Grown Single-Walled Carbon Nanotubes in the Helical Superstructure of Schizophyllan and Curdlan (*β*-1,3-Glucans)

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Abstract: We have found that single-chain schizophyllan and curdlan (s-SPG and s-curdlan, respectively) can dissolve as-grown and cut single-walled carbon nanotubes (ag-SWNTs and c-SWNTs, respectively) in aqueous solution. The vis−NIR spectra of the composites suggest that c-SWNTs are dissolved as a bundle, whereas ag-SWNTs exist as one or only a few pieces in the tubular hollow constructed by the helical structure inherent to these β-1,3-glucans. EDX and CLSM measurements and TEM observation established that the distribution map of these polysaccharides overlaps well with the image of SWNTs, indicating that these two components form a composite. Very interestingly, when c-SWNTs were dissolved with the aid of s-SPG or s-curdlan in water, a clear periodical structure with inclined stripes, as detected by AFM, appeared on the fibrous composite surface. Because this periodical structure has never been recognized for the composites with other water-soluble polymers, one can regard that s-SPG or s-curdlan wraps c-SWNTs constructing a helically twined structure. High-resolution TEM observation of an ag-SWNTs/s-SPG composite gave a clearer image in that two s-SPG chains twine one ag-SWNT and the helical motif is right-handed. When this sample was subjected to the AFM measurement, the composite showed the 2−3 nm height. This height implies that one piece of ag-SWNT is included in the s-SPGs helical structure. As a summary, it has been established that β-1,3-glucans such as s-SPG and s-curdlan not only dissolve SWNTs but also create a novel superstructure on the surface.

Introduction

Since the discovery of single-walled carbon nanotubes (SWNTs),[1] they have been expected to become a new potential source of functional materials.[2,3] However, their strong cohesive nature and poor solubility have caused researchers trouble for a long time as these properties seriously hamper efforts to obtain reproducible data. Because of the difficulty arising from these problems, reliable data on the SWNT superstructures have been obtained mainly from the samples extended on the surface, for example, by using an AFM technique.[4] One potential breakthrough in overcoming these problems is to derive them into the amphiphilic compounds by introducing appropriate solubilization groups either (1) into the SWNT end groups[5] or (2) into the side surface of SWNTs.[6] However, method (1) accompanies the difficulty in the product characterization, whereas in method (2) the electron conjugation system characteristic of SWNTs is inevitably damaged and the inherent functionalities may be partially reduced. An alternative method developed so far is to wrap SWNTs in water-soluble polymers. The polymers that are known to have such a wrapping ability are poly(vinylpyrrolidone),[7] amylose,[8] DNA,[9] peptides,[10] etc.[11] Among them, particularly interesting is amylose reported by Stoddart's group,[8] because (1) it has almost no light absorption in the UV−vis

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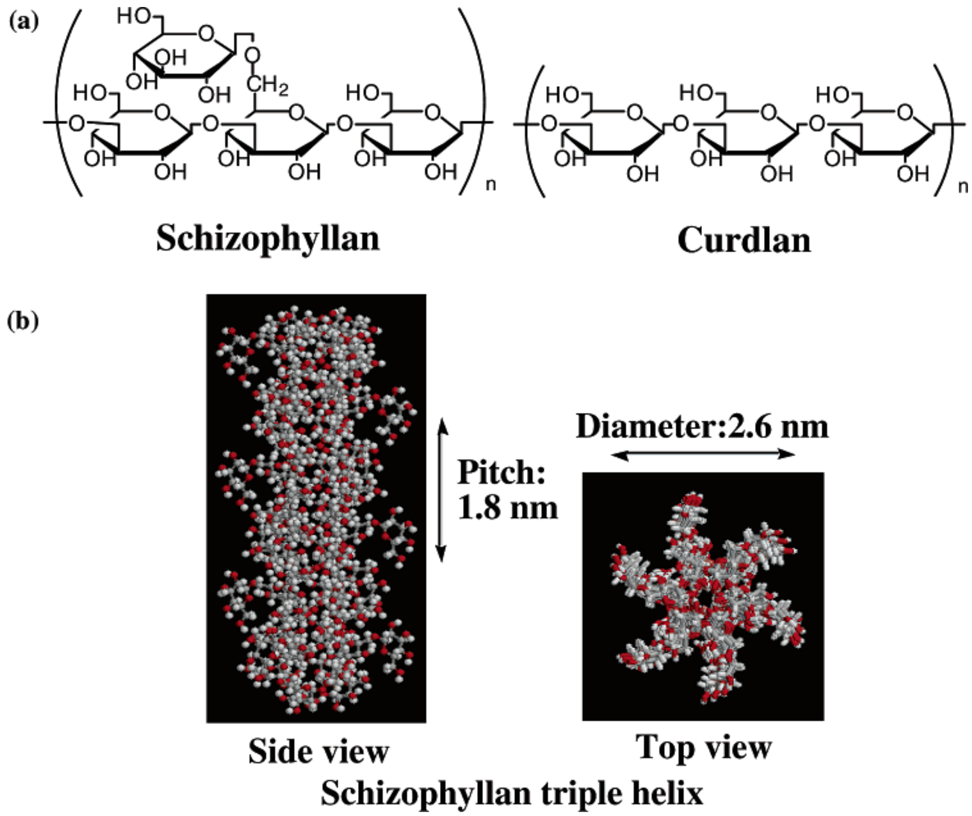

Figure 1. (a) Repeating unit of schizophyllan and curdlan; (b) a representive model of schizophyllan triple helix.

wavelength region, so that the dispersed composites are very suitable for photochemical experiments, (2) the sugar-coated surface of the composite should show biocompatibility, which would make medicinal applications possible, and (3) if the sugar group is recognized by proteins (such as lectins), one may construct novel SWNTs-based supramolecular network structures.[12] Furthermore, when amylose forms a helical structure to wrap SWNTs, the chirality information in amylose may be transduced to the π-conjugate systems in SWNTs.

Schizophyllan (SPG) is a natural polysaccharide produced by the fungus *Schizophyllum commune*, and its repeating unit consists of three β-(1−3) glucoses and one β-(1−6) glucose side-chain linked at every third main-chain glucose (Figure 1a).[13] SPG adopts a triple helix (t-SPG) in nature, which can be dissociated into a single chain (s-SPG) by dissolving in dimethyl sulfoxide (DMSO).[14] The s-SPG chain can retrieve the original triple helix by exchanging DMSO for water.[15] Recently, we found it has a novel solution property that when this renaturing process is carried out in the presence of certain polynucleotides, the resultant triple helix consists of two s-SPG chains and one nucleotide chain.[16] These preceding findings suggest that SPG

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may show a unique affinity to form stable complexes with one-dimensionally extended compounds.

It is known that in the glucose unit the 2-OH side is a hydrophobic face whereas the 6-OH side is a hydrophilic face.[17a] Because the triple-stranded helical structure of SPG is stabilized by the hydrogen-bonding network among the 2-OH groups formed inside the helical column,[17b,c] one may presume that the inside hollow is considerably hydrophobic, like the inside cavity of cyclodextrins (Figure 1b).[18] In fact, the driving force for SPG chains to reconstruct the triple strand from the single strand is considered to be a hydrophobic force in addition to the hydrogen-bonding force.[15] It thus occurred to us that SWNTs might be entrapped, because of the hydrophobic interaction, in the inside hollow of the SPG helical structure. In fact, it is known that I_2 and surfactants with long alkyl chains are included inside the helical structure of starch.[19] Moreover, Dodziuk et al. found that η-cyclodextrin consisting of 12 glucose units forms a pseudo-rotaxane-like complex with a SWNT.[20] We here report that s-SPG and s-curdlan [a single chain of curdlan (β-1,3-glucan without a side-chain glucose): Figure 1a] are capable of wrapping not only SWNTs cut to 1−2 μm length (c-SWNTs) but also as-grown SWNTs (ag-SWNTs: in some cases, one-piece of ag-SWNT), and the "periodical" stripes arising from the twining SPG chains are clearly observable by AFM and TEM, reflecting the strong helix-forming nature of the β-1,3-glucan main chain.[21] We consider that this is the first example that demonstrates a visual image indicating how the water-soluble polymer wraps the SWNT.

Results and Discussion

Dissolution of c-SWNTs by Wrapping with s-SPG and s-Curdlan. Vis−NIR and Raman Spectroscopic Measurements and TEM Observation. We first studied this system using c-SWNTs because the handling is easier than that of ag-SWNTs. The preparation of c-SWNTs was described previously.[4] The samples for the spectroscopic measurements were prepared by mixing an aqueous solution of dispersed c-SWNTs with a DMSO solution containing s-SPG or s-curdlan (for details, see the Experimental Section). The presence of c-SWNTs in the composite was evidenced by the measurements of vis−NIR and Raman spectroscopy. Figure 2 shows a vis−NIR spectrum of the composite. The characteristic absorption band is seen in both the 800 and the 1400 nm regions, which are basically similar to those of the reported spectra.[9c,11c] As the peaks are significantly broadened, one can presume that c-SWNTs are aggregated into bundles to some extent. It is known that c-SWNTs have characteristic Raman peaks at around 190 and 1590 cm^{-1}.[11a,22] We have confirmed that c-SWNTs/s-SPG, c-SWNTs/s-curdlan (M_w = 9500), and c-SWNTs/s-curdlan (M_w = 33 000) composites all give the peaks at 188 and 1591 cm^{-1} (Figure 3). As a reference, we prepared a c-SWNTs/amylose composite according to Stoddart's method.[8a] The Raman spectrum arising from the c-SWNTs/amylose composite is very similar to those obtained in the present study.

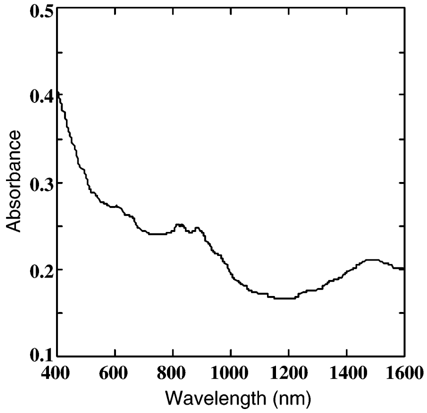

Figure 2. Vis−NIR spectrum of c-SWNT/s-SPG solution: D_2O, 1 cm cell, room temperature. The spectrum of c-SWNT itself was almost the same as that of the c-SWNT/s-SPG composite shown above.

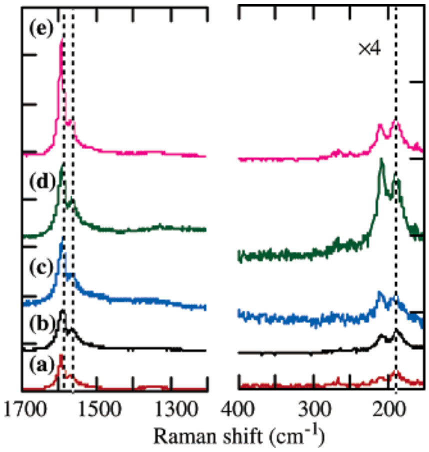

Figure 3. Raman spectra of (a) c-SWNTs, (b) c-SWNTs/s-curdlan (M_w = 9500) composite, (c) c-SWNTs/s-SPG composite, (d) c-SWNTs/s-curdlan (M_w = 33 300) composite, and (e) c-SWNTs/amylose composite.

TEM images of c-SWNTs themselves and their composites with s-SPG or s-curdlan (M_w = 33 000) are shown in Figure 4. Figure 4a indicates that the length of c-SWNTs is several micrometers, which is comparable to the length estimated by AFM. However, the diameter size is somewhat larger, indicating that they are easily aggregated into the bundles in the absence of water-soluble polymers acting as solubilizers. In the presence of s-SPG or s-curdlan (M_w = 33 000), on the other hand, c-SWNTs stably exist as slender fibrils, suggesting that the

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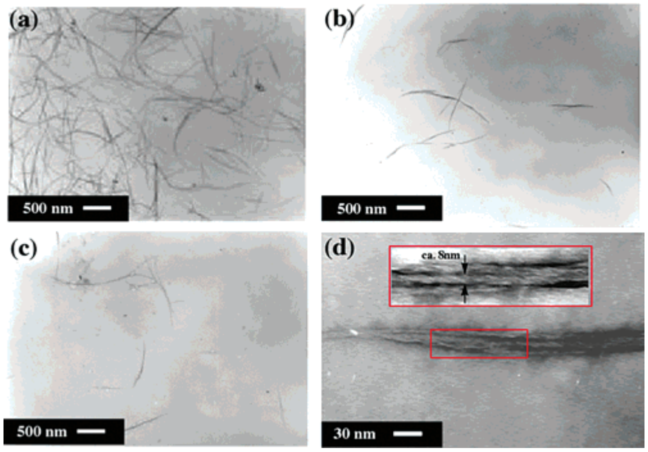

Figure 4. TEM images of (a) c-SWNTs, (b) c-SWNTs/s-SPG composite, (c) c-SWNTs/s-curdlan composite, and (d) a magnified picture of (b). (a)−(c) were taken without staining, whereas (d) was taken after staining with 2.0 wt % phosphotungstic acid.

wrapping by polysaccharides suppresses the aggregation of c-SWNTs (Figure 4b and c). To obtain a clearer image of the composite, we magnified one fibril after staining with phosphotungstic acid (Figure 4d). Interestingly, one can confirm that this fibril consists of a few bundles with ca. 8 nm diameter and each bundle is decorated by stripes. From the inclination of these stripes (right up, left down), one can presume that these bundles have a right-handed helical motif. These results show that s-SPG chains twine around a c-SWNT bundle, creating a periodical helical pattern on the surface of the bundle. Here, one may consider that the diameter of the composite is larger than that of t-SPG itself. It is rather natural, however, that s-SPG includes somewhat larger guest polymers, because the main chain of β-1,3-glucans is not so rigid as cyclodextrins and can take various higher-order structures. The result indicates that the cavity size of SPG can be changed to accommodate the size of entrapped guest polymers in an induced-fit manner. As reference experiments, we prepared the samples of c-SWNTs itself and t-SPG itself according to the same treatment. However, we could not recognize this kind of stripe at all.

EDX-TEM Observation. The presence of polysaccharides in the composite was evidenced by EDX (energy-dispersive X-ray spectroscopy). Figure 5a−c shows a TEM image, an EDX spectrum, and an EDX line scan profile, respectively, of c-SWNTs themselves. As is seen in Figure 5b and c, a small amount of oxygen is detected in addition to a large amount of carbon. Presumably, this peak is assignable to the functional groups such as OH, COOH, etc., produced by the cutting treatment of ag-SWNTs by oxidation. The C/O ratio (wt/wt) estimated by this elemental analysis is 94.9/2.1. On the other hand, Figure 5d−f for the c-SWNTs/s-SPG composite shows the presence of saccharide-based oxygen atoms, the C/O ratio (wt/wt) being 68.1/18.3. The decreased C/O ratio and the fact that the oxygen intensity is not distributed beyond the carbon intensity imply that the thin oxygen-containing layer of s-SPG chains is constructed around the bundle of c-SWNTs.[23]

CLSM and TEM Observation. Formation of the composite was further confirmed by a confocal laser scanning microscope (CLSM) using an s-curdlan covalently linked to fluorescein (s-curdlan-F): the average molecular weight of the s-curdlan is ca. 8100 with $M_w/M_n = 1.2$. This s-curdlan-F gave blue fluorescence under UV light irradiation (Figure 6a), whereas c-SWNTs appeared as black shadows under the optical microscopic observation (Figure 6b). As shown in Figure 6c, the blue fluorescence in Figure 6a and the black shadows in Figure 6b were perfectly overlapped, indicating that c-SWNTs and s-curdlan-F coexist in the same domain. When the red circles in Figure 6c were photoexcited at 364 nm, they gave fluorescence spectra having an emission maximum at 527 nm (Figure 6d). The result confirms that this blue color is emitted from the excited state of fluorescein.[24]

To obtain unequivocal evidence that the polymeric chains of β-1,3-glucan polysaccharides really construct a periodical helical superstructure around the bundle of c-SWNTs, we observed these composite fibrils by AFM. As shown in Figure 7a, the surface of c-SWNTs themselves did not give any specific pattern. We also prepared c-SWNTs solutions dispersed by amylose or poly(vinylpyrrolidone), but their AFM images are similar to that in Figure 7a. This implies that, even though these water-soluble polymers may wrap c-SWNTs to dissolve them into water, the periodical structure is not constructed on the c-SWNTs surface in their wrapping process. On the other hand, when a DMSO solution of s-SPG was cast on mica, we could observe a fine polymeric network structure and the height of one chain was estimated to be ca. 1 nm (Figure 8b). Very interesting, when c-SWNTs were dispersed with the aid of s-SPG or s-curdlan ($M_w = 33\,000$) into water, the surface of the fibrils showed a periodical structure with inclined stripes (Figure 7b−d). In a magnified picture of these fibrils (Figure 7d), one can clearly recognize that the well-ordered, periodical structure is constructed on the surface. Here, it should be noted that the periodical structure of t-SPG could not be visually recognized under the same conditions due to its small diameter. We also confirmed the fact that the scanning direction did not affect the periodical wave patterns at all. The findings imply that this novel periodical structure is observable only for the β-1,3-glucan polysaccharides because of their strong helix-forming nature. As far as we know, this is the first example in which construction of the superstructure on the SWNTs surface is visually confirmed.[25] Subsequently, the periodical interval in the helical stripes was estimated by AFM. It is confirmed from scanning along a fibril that the mountain and the valley appear periodically, like the teeth of a saw, at every 16 nm interval (Figure 8a). On the other hand, when the mica surface beside the composite was scanned along the fibril, we could not observe any periodical pattern at all. It is also confirmed from AFM that most composites have ca. 10 nm height, indicating that β-1,3-glucan polysaccharides twine around a bundle of c-SWNTs.

To confirm the fact that the periodical wave patterns are not affected by the AFM tip, we observed the composite fibrils by SEM.[16c] The results are shown in Figure 9. One can recognize

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[23] To obtain the elemental analysis data with a good S/N ratio, we had to select somewhat larger bundles that would be formed during the drying process, otherwise we could not obtain the reliable elemental analysis data.

[24] It should be noted that the CLSM image reflects the fluorescence of the composite, so that the size (or width) of the composite cannot be estimated by this fluorescence method.

[25] Synthetic polymers that are covalently attached to carbon nanotubes tend to form regular structures on the carbon nanotube template. See: (a) Czerw, R.; Guo, Z.; Ajayan, P. M.; Sun, Y.-P.; Carroll, D. L. *Nano Lett.* **2001**, *1*, 423. For a related paper, see: (b) Murphy, R.; Coleman, J. N.; Cadek, M.; McCarthy, B.; Bent, M.; Drury, A.; Barklie, R. C.; Blau, W. J. *J. Phys. Chem. B* **2002**, *106*, 3087.

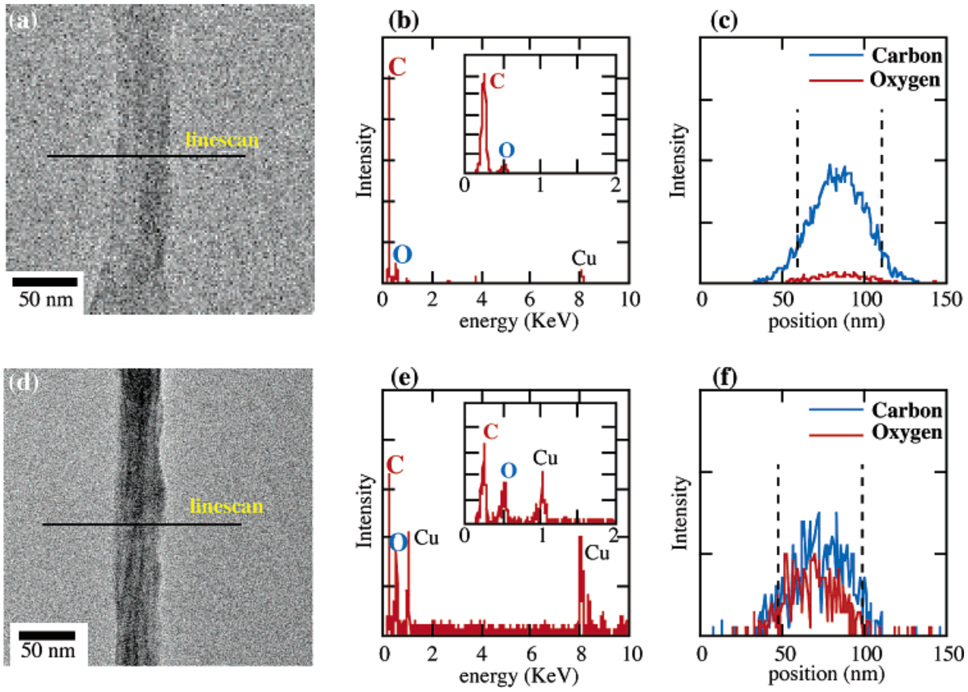

Figure 5. TEM images, elemental analysis based on EDX, and EDX line profiles: (a)−(c) for c-SWNTs and (d)−(f) for c-SWNTs/s-SPG composite.

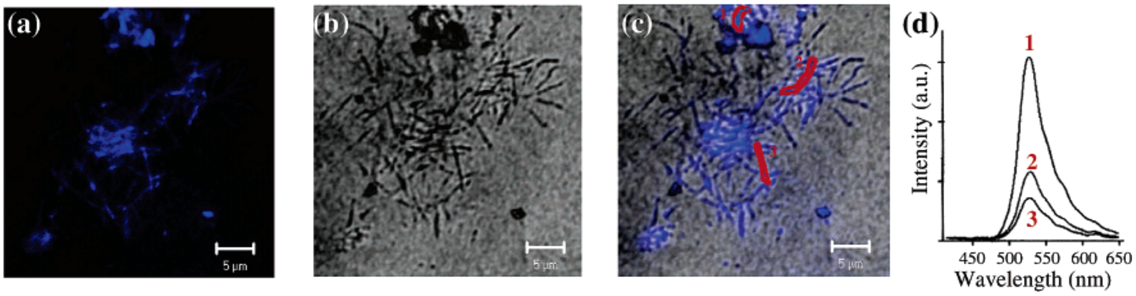

Figure 6. CLSM images of c-SWNTs/s-curdlan-F composite obtained by (a) fluorescence microscope, (b) optical microscope, (c) overlap of (a) and (b), and (d) fluorescence spectra for excitation of the red circles in (c) at 364 nm (the numbers of spectra in (d) correspond to those of red circles in (c)).

from Figure 9 that the diameter of the composite is 20−30 nm and the periodical structure is constructed on the c-SWNT surface. The thickness of the platinum coating is estimated to be 10 nm from the coating time and current. Therefore, the actual diameter of the composite is estimated to be 10−20 nm, which is consistent with the results obtained from TEM and AFM.

As a summary of the foregoing findings, one can now conclude that (1) the fibrous structures observable with microscopes are composites formed from c-SWNTs and s-SPG (or s-curdlan) and (2) the helical twining pattern constructed from these polysaccharide chains is visually seized by AFM, TEM, and SEM. s-SPG and s-curdlan possessing a β-1,3-glucan main-chain structure are able to dissolve "cut" c-SWNTs in aqueous solution, but "cut" c-SWNTs can be dispersed in aqueous solution even in the absence of water-soluble polymers, although they tend to aggregate by themselves into the bundle. To further evaluate the solubilization ability of these polysaccharides, we applied s-SPG to ag-SWNTs that cannot be dissolved without the aid of surfactants or water-soluble polymers. As explained below, s-SPG showed a remarkable solubilization power even for ag-SWNTs. s-Curdlan also showed a similar solubilization power, but the ability is somewhat inferior to that of s-SPG because of its poor solubility in water. Here, we mainly report the data we obtained from the ag-SWNTs/s-SPG composite.

Dissolution of ag-SWNTs by Wrapping with s-SPG. Procedure for Dissolution of ag-SWNTs. A DMSO solution containing ag-SWNTs and s-SPG was sonicated in a water bath (the details are described in the Experimental Section). The mixed solution gradually became dark as the sonication time progressed. This mixture was diluted with water under vigorous stirring. The resultant mixture was subjected to centrifugation, the ag-SWNTs/s-SPG composites being recovered as precipitate. This process was repeated five times. The AFM measurements showed that the precipitate consists only of the ag-SWNTs/s-SPG composites while the supernatant contains uncomplexed s-SPG, amorphous carbons (impurity in ag-SWNTs), etc. This implies that this dissolution treatment is useful for purification of ag-SWNTs. The maximum concentration of ag-SWNTs dissolved in water by this procedure was 2.5 mg mL^{-1}.[26] We confirmed that if the resultant solution is diluted with water (lower than 0.5 mg mL^{-1}), it can exist as a clear solution over 1 month. We performed the same experi-

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(26) The concentration was determined using a reported extinction coefficient. See: Bahr, J. L.; Mickelson, E. T.; Bronikowski, M. J.; Smalley, R. E.; Tour, J. M. *Chem. Commun.* **2001**, 193.

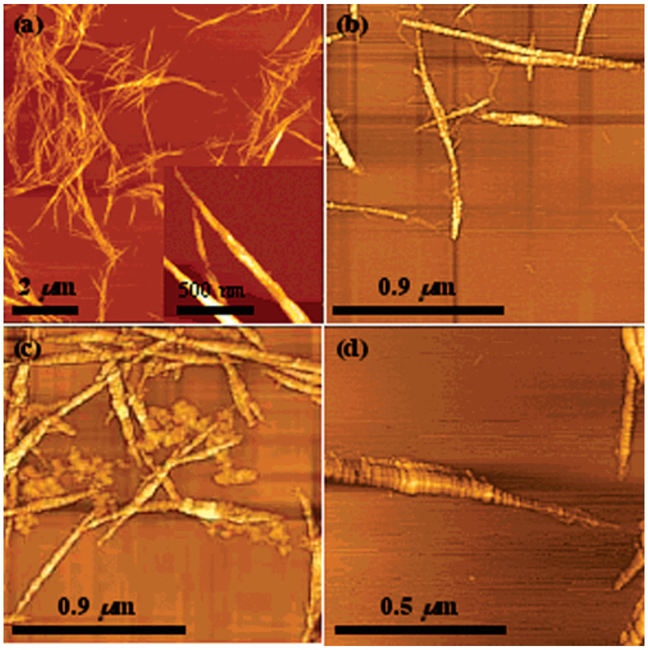

Figure 7. AFM images of (a) c-SWNTs, (b) c-SWNTs/s-SPG composite, (c) c-SWNTs/s-curdlan composite, and (d) a magnified picture of fibrils in s-SWNTs/s-SPG composite. In (c), amorphous mass observed around the fibrous composite is attributed to curdlan, which could not form the composite with c-SWNTs. The same AFM image was obtained from curdlan itself. The difference between (b) and (d) is caused by the poor water solubility of curdlan.

ments with amylose, dextran, pulluran, or starch, but none of them could disperse ag-SWNTs into water. The difference clearly indicates the superiority of β-1,3-glucan polysaccharides as a one-dimensional host for ag-SWNTs over other polysaccharides.

It is known that DMSO and alkaline aqueous solutions are good solvents for s-SPG.[14,15] It thus occurred to us that the ag-SWNTs/s-SPG composite may be dissociated into each component in these solvents. As shown by pictures in Figure 10, ag-SWNTs were immediately precipitated when these solvents were added (the concentration in the final solutions was 50 vol %). The AFM measurement proved that the supernatant contains only dissociated s-SPG. We consider that this process will be useful for recovering "pure" ag-SWNTs from the composite.

Estimation of Dispersity by Vis−NIR Spectroscopy. ag-SWNTs were dispersed into D_2O with the aid of s-SPG, and the mixture was subjected to vis−NIR measurements (Figure 11). As the peaks have become much sharper than those of bundled c-SWNTs (Figure 2), one can clearly assign them to characteristic bands in ag-SWNTs.[27] The result suggests that one or a few pieces of ag-SWNTs are included in the SPG helical structure. Smalley et al. raised the sharpness of the NIR peaks as evidence for discrete dispersion of SWNTs: for example, they proposed that SWNTs dispersed by sodium dodecyl sulfate (SDS) micelles exist as one piece in the micelle.[27c] Because of the difference in the ag-SWNTs source (see Experimental Section), however, our vis−NIR spectral data cannot be compared with theirs directly. We thus prepared the SDS solution containing our ag-SWNTs by ourselves according to Smalley's procedure. As a result, we could confirm that the peak sharpness of the vis−NIR spectrum obtained from the s-SPG-dispersed system is almost the same as that obtained from the SDS-dispersed system. The finding supports the view that in the present s-SPG system, ag-SWNTs are well-dispersed as one or only a few pieces.

High-Resolution TEM and Three-Dimensional TEM Observation. The ag-SWNTs/s-SPG composite was characterized by high-resolution TEM (HRTEM).[28] From Figure 12a, the composites are visually recognized to be a very small, fibrous structure but not a bundled structure. Figure 12b and c shows the enlarged pictures. From Figure 12c, one can clearly recognize that two s-SPG chains twine around one ag-SWNT with 1.5 nm diameter. It is also seen from Figure 12c that the helical motif is a right-handed one and the helical pitch of one chain is ca. 10 nm (in Figure 12d, the original image is Fourier filtered to enhance the wrapping pattern more clearly). These TEM images provide "decisive" evidence that one piece of ag-SWNTs is wrapped by s-SPG chains.[29]

Furthermore, we succeeded in taking a three-dimensional (3-D) image of this picture.[30] The four selected still pictures are shown in Figure 13, and the movie is deposited as Supporting Information. In Figure 13, each 3-D image corresponds to the clockwise rotation of the "Y" shape structure in Figure 12b with 45°, 135°, 225°, and 315° along a perpendicular axis. In these 3-D images, one can see that ag-SWNT appears as a white fine fibril (for example, right branch in the 45° rotating image) and a SPG thin layer wraps the fibril. From these 3-D TEM images, one can confirm that the s-SPG chains exist as a uniform layer around one piece of ag-SWNTs, and the composite structure can be viewed from any angle. This result strongly supports the fact that s-SPG wraps one piece of ag-SWNT.

AFM Observation of ag-SWNTs/s-SPG Composites. The AFM images of the sample obtained by casting the ag-SWNTs/s-SPG composite solution on mica are shown in Figure 14. It is seen from Figure 14a that the fibrous composites are well dispersed on the mica surface and most of them have a 2−3 nm height. This result is supported more quantitatively by the histogram (Figure 14b): the height distribution is very

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(27) (a) Ausman, K. D.; Piner, R.; Lourie, O.; Ruoff, R. S. *J. Phys. Chem. B* **2000**, *104*, 8911. (b) Chiang, I. W.; Brinson, B. E.; Huang, A. Y.; Willis, P. A.; Bronikowski, M. J.; Margrave, J. L.; Smalley, R. E.; Hauge, R. H. *J. Phys. Chem B* **2001**, *105*, 8297. (c) O'Connell, M. J.; Bachilo, S. M.; Huffman, C. B.; Moore, V. C.; Strano, M. S.; Haroz, E. H.; Rialon, K. L.; Boul, P. J.; Noon, W. H.; Kittrell, K. L.; Ma, J.; Hauge, R. H.; Weisman, R. B.; Smalley, R. E. *Science* **2002**, *297*, 593.

(28) (a) The electron conductivity of carbon nanotubes plays an important role in shielding the sample from electron beam damage. See: (b) Tsang, S. C.; Guo, Z.; Chen, Y. K.; Green, M. L. H.; Hill, H. A. O.; Hambley, T. W.; Sadler, P. J. *Angew. Chem., Int. Ed. Engl.* **1997**, *36*, 2197. (c) Guo, Z.; Sadler, P. J.; Tsang, S. C. *Adv. Mater.* **1998**, *10*, 701. For a recent HRTEM review, see: (d) Thomas, J. M.; Midgley, P. A. *Chem. Commun.* **2004**, 1253.

(29) Recently, Coleman and Ferreira presented a simple and general model that describes the ordered assembly of polymer strands on nanotube surfaces, the energetically favorable coiling angles being estimated to be 48°−70°: Coleman, J. N.; Ferreira, M. S. *Appl. Phys. Lett.* **2004**, *84*, 798. This theoretical approach is partly complementary to our present results, but their single-chain wrapping system cannot be directly compared to our two-chains wrapping system.

(30) Electron tomography (ET) is a useful technique for reconstructing an object from a series of projections acquired by TEM. Recent development of fully digitized and automated TEM has enabled us to achieve the three-dimensional ET, not only to determine the size and distribution of objects but also to provide information about the morphology of them in the nanometer order. Three-dimensional structure was reconstructed by processing a series of projections by combination of IMOD and Amira. See: (a) Frank, J. *Three-dimensional Electron Microscopy of Macromolecular Assemblies*; Academic Press: San Diego, CA, 1996. (b) Midgley, P.; Weyland, M. *Ultramicroscopy* **2003**, *96*, 413. (c) Kermer, J. R.; Mastronarde, D. N.; McIntosh, J. R. *J. Struct. Biol.* **1996**, *116*, 71.

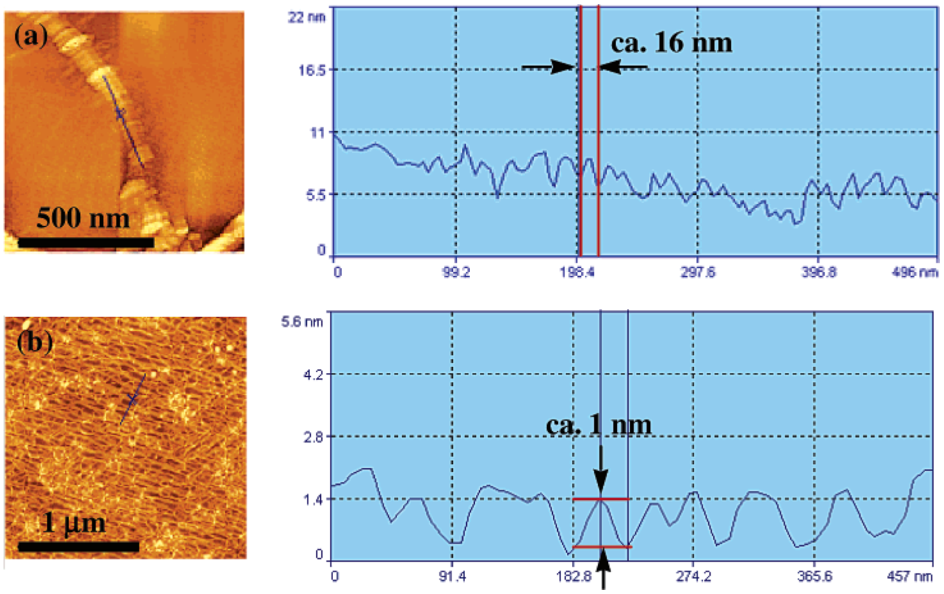

Figure 8. AFM images and scanning profiles of vertical sectional plans of (a) c-SWNTs/s-SPG composite, and (b) s-SPG. AFM tip was scanned along the blue lines.

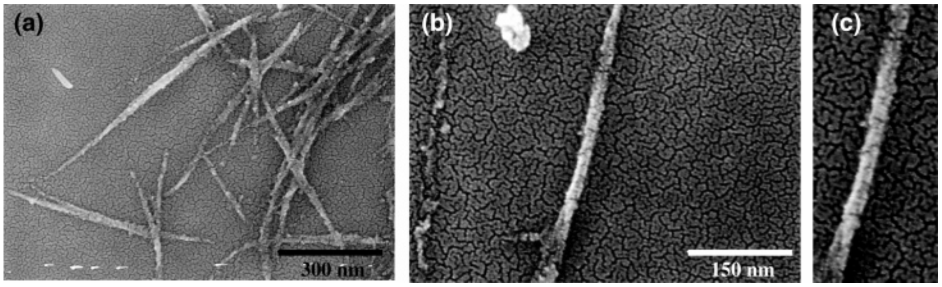

Figure 9. (a,b) SEM image of c-SWNTs/s-SPG composite; (c) magnified picture of (b).

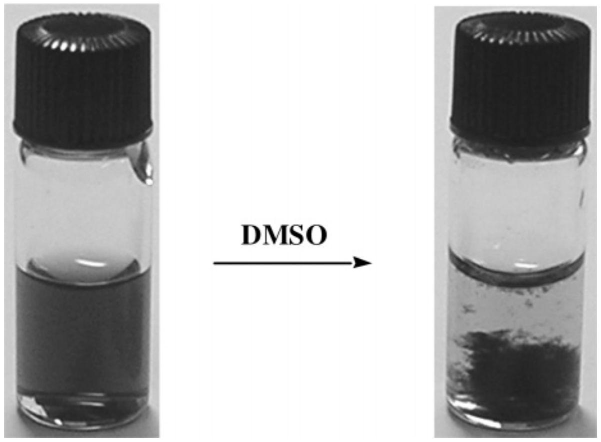

Figure 10. Picture of aqueous ag-SWNTs/s-SPG composite solution (left) and that taken after addition of DMSO (right): the final solution contains 50 vol % DMSO. Addition of aqueous 1.0 M NaOH solution results in a similar precipitate of ag-SWNTs.

narrow, and the peak maximum appears at 2.5 nm. As the diameter ofag-SWNT is 1.5 nm and the size of the s-SPG chain is estimated to be ca. 1 nm (Figure 8b), the 2.5 nm height implies that the composite consists, mostly, of one piece of ag-SWNT wrapped by the s-SPG chains. The size of these composites is so small that the stripe pattern as observed for the c-SWNTs/s-SPG composites (Figure 7) is not visually recognized.

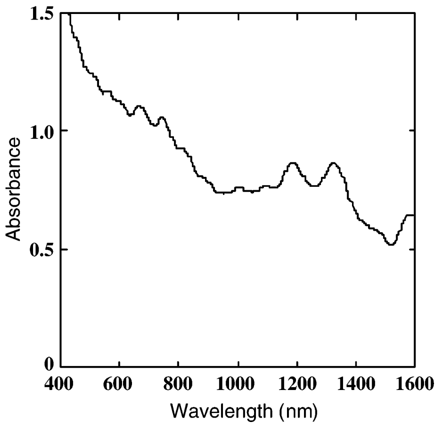

Figure 11. Vis−NIR spectrum of ag-SWNTs/s-SPG solution: D_2O, 1 cm cell, room temperature.

However, when the surface of the composite was scanned along the fibril, a periodical wave pattern with a ca. 12 nm interval appeared. This periodical wave pattern is similar to that observed in Figure 8a. One may consider, therefore, that this arises from the s-SPG chains helically wrapping ag-SWNT.

Conclusion

Various spectroscopic measurements and microscopic observations conducted herein consistently support the view that the water-soluble nanofibers are comprised of SWNTs and

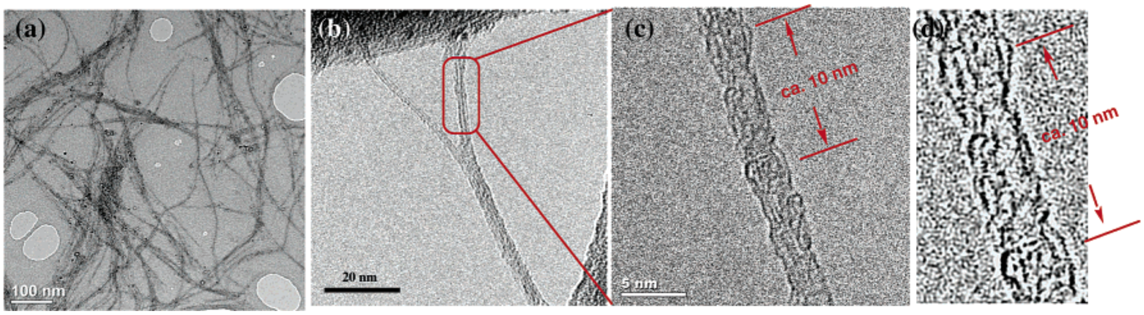

Figure 12. (a) TEM image of ag-SWNT/s-SPG composite, and (b,c) its magnified picture. (d) The original image of (c) was Fourier filtered to enhance the contrast of the composite.

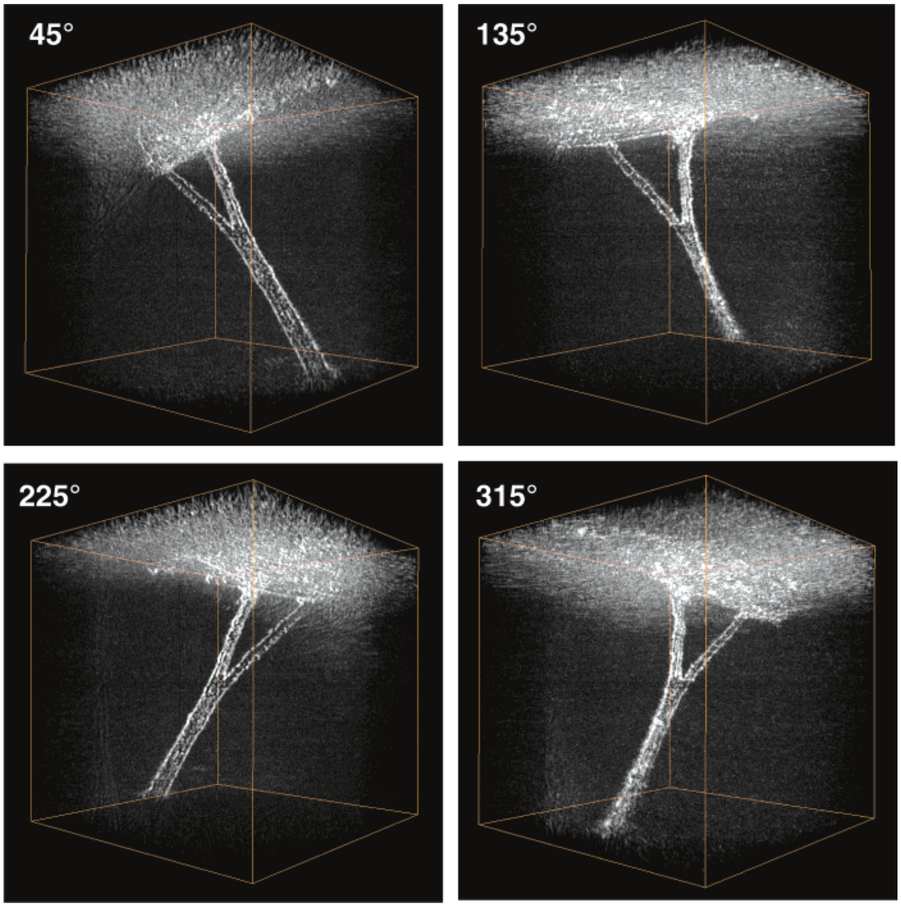

Figure 13. Reconstructed 3-D images of ag-SWNTs/s-SPG composite, viewed from different orientations. These correspond to the clockwise rotation images of the 2-D image in Figure 12b, rotating around a perpendicular axis with 45°, 135°, 225°, and 315°, respectively.

β-1,3-gulucan polysaccharides. Particularly interesting are the findings that (1) the periodical stripe structure, which stems from a helical wrapping mode characteristic of β-1,3-glucans, is confirmed, (2) the composite with ag-SWNTs contains mainly one piece of ag-SWNT, and (3) the “important” picture that clearly evidences that two s-SPG chains helically wrap one piece of ag-SWNT and the helicity is right-handed has been taken. These lines of clear image on the composites are obtained because of the helix-forming nature of β-1,3-glucans.

So far, it has not been so easy to prepare a stable composite including one piece of SWNT isolated from each other. The present paper shows that the polymer wrapping with β-1,3-glucan polysaccharides is one promising method. To introduce functional groups into SWNTs, covalent-bond formation has been mainly utilized, which partially damages the π-conjugate system. The present paper suggests that such functional groups can be introduced into the β-1,3-glucan polysaccharides, without damaging the π-conjugate system. Furthermore, the presence of the periodical structure on the composite surface implies that one can combine the present system with the concept of supramolecular or self-assembly chemistry. We believe, therefore, that the present system has a broad future potential both in chemical modification and in physical characterization of SWNTs.

Experimental Section

Materials. SPG was kindly supplied by Taito Co., Japan. The molecular weight and the number of repeating units were evaluated to be 1.5 × 10^5 and 231, respectively. Other polysaccharides used here were purchased from Tokyo Kasei Kougyou, Co. Native curdlan

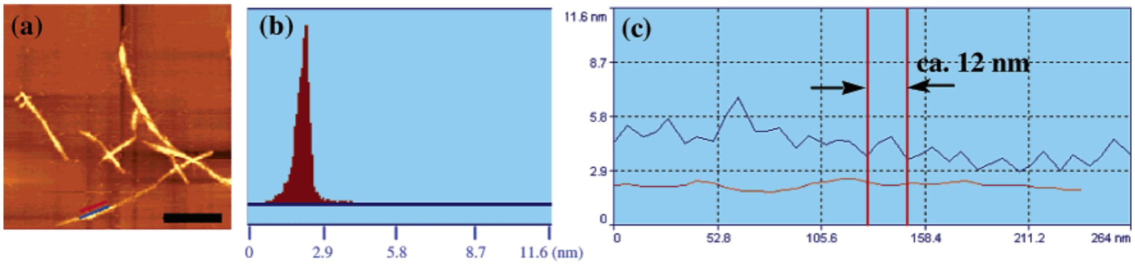

Figure 14. (a) AFM images of ag-SWNTs/s-SPG composite (bar = 500 nm), (b) height distribution of the composites, and (c) scanning profiles of vertical sectional plans of the composites (blue) and the mica surface (red). AFM tip was scanned along the blue and red lines in (a).

(M_w = 1 200 000) is scarcely soluble in water, but when the M_w is reduced, it becomes moderately water-soluble.[31] DMSO was obtained from Kishida Chemical Co. and was used for all experiments. D_2O was purchased from Wako Pure Chemical Industries and was used for vis−NIR measurements. SWNTs produced by the HiPco (high-pressure decomposition of carbon monoxide) process were obtained from Carbon Nanotechnologies, Inc. ag-SWNTs were cut into the appropriate length as described previously.[4] The length as estimated by AFM was between 1 and 2 μm.

Preparation of c-SWNTs/Polysaccharide Composites. An aqueous solution of dispered c-SWNTs (250 μL) was mixed with 50 μL of DMSO solution containing s-SPG (5.0 mg mL^{-1}, M_w = 150 000). At this stage, the water/DMSO mixed solutions contained 300−600 μg mL^{-1} of c-SWNTs and 830 μg mL^{-1} of s-SPG, and the composition of water/DMSO (v/v) was 84/16 (v/v). After being left for 2 days at room temperature, the mixture was treated with a centrifuge (7000 rpm) for 1 h and the supernatant that contains uncomplexed s-SPG was pipetted off. The precipitated c-SWNTs/s-SPG composite was then dispersed into water (200 μL). After repeating this centrifuge process five times, the clear aqueous solution was obtained. The c-SWNTs/s-curdlan (M_w = 9500 and 33 300) composites were prepared by the same procedure. Two control samples using amylose (M_w = 15 000) were prepared according to reported procedures.[8a,b]

Preparation of ag-SWNTs/Polysaccharide Composites. The procedure used for dissolution of ag-SWNTs is as follows. HiPco SWNTs (0.5 mg) were weighted in a sample tube. To the tube, 2 mL of DMSO was added, and SWNTs were dispersed by sonication (50 W, 20 kHz) for 3 h using a probe type sonicator with the sample immersed in a water bath. The sonicator tip was placed into the sample by one-third of the distance from the surface. To the resultant dispersion, 500 μL of DMSO solution containing s-SPG (5.0 mg mL^{-1}, M_w = 150 000) was added, and the mixture was sonicated for 10 min in a water bath. The mixture was gradually diluted with water (2.5 mL) extending over 10 min under sonication (50 W, 20 kHz). The sonicated mixture was added to 35 mL of water to give a homogeneous black solution. The final composition of water/DMSO (v/v) was 94/6 (v/v). The solution was left for 2 days at room temperature to complete the renaturating process of s-SPG. The solution was then subjected to centrifugation at 12 000 rpm for 2 h. After the supernatant was removed, the precipitate was dispersed into 2.0 mL of water again (for vis−NIR sample preparation, D_2O was used instead of water at this process). Repeating this centrifuge process five times, uncomplexed s-SPG and amorphous carbons were pipetted off as a supernatant, and the clear black aqueous solution was obtained. The resultant solution was clear for over 1 month, unless it was of extremely high concentration. In the absence of SPG, on the other hand, ag-SWNTs were scarcely dissolved under the same treatment. The ag-SWNTs/s-curdlan composite was prepared by the same procedure. The control sample using SDS was prepared according to a reported procedure.[27c]

Synthesis of s-Curdlan-F. s-Curdlan covalently linked to fluorescein (s-curdlan-F) was synthesized according to the following procedure: curdlan (50 mg, M_w = 8100) was dissolved in DMSO (5 mL). To the DMSO solution, 13 mg (excess amount) of fluorescein-5-thiosemicarbazide was added, and the DMSO solution was stirred for 5 days at room temperature. The resultant solution was subjected to dialysis with a cellulose membrane (MWCO = 3500). Uncomplexed fluorescein-5-thiosemicarbazide was removed completely in this process. Frozen-dry treatment of the aqueous solution afforded the yellow powder. The obtained s-curdlan-F was used as a DMSO solution (5 mg mL^{-1}). The c-SWNTs/s-curdlan-F composite was prepared according to the same procedure described above.

Vis−NIR Absorption Spectrometry. Vis−NIR spectroscopic studies were performed on a SHIMADZU UV-3100 spectrophotometer.

Raman Spectrometry. Raman spectra of c-SWNTs/polysaccharide composites were obtained using a JASCO NRS-2000 laser Raman spectrometer.

Transmission Electron Microscopy (TEM) and High-Resolution TEM (HRTEM). TEM and HRTEM images were acquired using a JEOL TEM-2010 (accelerate voltage 120 kV) and a TECNAI-20, FEI (accelerate voltage 200 kV), respectively. c-SWNTs/s-SPG solution was placed on a copper TEM grid with a holey carbon support film. The TEM grid was dried under reduced pressure for 6 h before TEM observation.

Scanning Electron Microscopy (SEM). SEM studies were carried out on a Hitachi S-5000. c-SWNTs/s-SPG composite was shielded with platin. The accelerating voltage was 25.0 V, and the emission current was 10 μA. The sample was cast on mica and dried for 6 h under reduced pressure before SEM observation.

Energy Dispersive X-ray Spectroscopy (EDX). EDX was used to detect oxygen from s-SPG and to confirm the presence of s-SPG around c-SWNTs. c-SWNTs/s-SPG solution and c-SWNTs dispersion were placed on a copper TEM grid with a holey carbon support film, respectively. c-SWNTs were used to obtain a reference EDX line profile. Although we tried to use ag-SWNTs instead of c-SWNTs, it was difficult to cast them on a grid due to their poor solubility into water and strong cohesive nature. Therefore, c-SWNTs and their composite were examined by EDX here. EDX spectra and EDX line scan profiles were acquired from a holey place where c-SWNTs or c-SWNTs/s-SPG composite bridge over the hole. EDX spectra and EDX line scan profiles were obtained using a TECNAI-20, FEI.

Confocal Laser Scanning Microscope (CLSM). CLSM was used for taking the fluorescence image of s-curdlan-F. One drop of c-SWNTs/s-curdlan-F aqueous solution was sandwiched between cover glasses and fixed by an adhesive. CLSM images were taken via a Carl Zeiss LSM 510. Excitation wavelength was 364 nm.

Atomic Force Microscopy (AFM). AFM images were acquired in air using a Topo METRIX SPM 2100 (noncontact mode). The sample was cast on mica and dried for 6 h under reduced pressure before AFM observation.

Three-Dimensional TEM (3D-TEM). 3-D TEM images were taken using a TECNAI-20, FEI. The TEM parameters were controlled during the acquisition of a tilted series of projections by a cooled slow-scan

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(31) Koumoto, K.; Kimura, T.; Kobayashi, H.; Sakurai, K.; Shinkai, S. *Chem. Lett.* **2001**, 908.

charge-coupled device (CCD) camera for 3-D electron tomography (3-D ET) by fully digitized and automated TEM. A series of tilted projections was acquired for this experiment from −60° to 60°, with an image recorded every 2° giving a total of 60 images. The exposure time is as short as 1.0 s to reduce the effect of irradiation damage on the samples. Once the acquisition of a tilt series was complete, the data were transferred to a workstation for fine-tuning of the alignments and for 3-D reconstruction. These data were spatially aligned using a cross-correlation algorithm from IMOD software, and a 3-D reconstruction was achieved using a weighted back-projection of consecutive 2D slices, from AMIRA 3.0.[30c]

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Supporting Information Available: Two movies of the 3-D TEM image. This material is available free of charge via the Internet at http://pubs.acs.org.

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