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# Preparation and Characterization of Grafted Collagen-Multiwalled Carbon Nanotubes Composites

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This paper describes a new class of composite materials designed by combining multiwalled carbon nanotubes (MWCNTs) and grafted collagen matrix. These materials show high mechanical capabilities by taking advantage of the favorable mechanical characteristics of MWCNTs. Furthermore, doping carbon nanotubes into grafted collagen matrix results in a substantial improvement of thermal stability and infrared emissivity. Thus these materials possess potential applications in some fields such as biomedicine and infrared camouflage.

**Keywords:** Composites, Grafted Collagen, Multiwalled Carbon Nanotubes, Mechanical Capability, Thermal Stability, Infrared Emissivity, Infrared Camouflage.

RESEARCH ARTICLE

## 1. INTRODUCTION

Since the discovery of carbon nanotubes (CNTs) in 1991 by Iijima,[1] many efforts have been devoted to revealing their unique structural, electrical, mechanical, and chemical properties.[2–5] Recently, extensive research on CNTs has concentrated on incorporation of CNTs into polymeric matrix for improving electrical conductivity,[6] optical character,[7] mechanical strength,[8] and other characters,[9] which expand the applications of polymer materials in many fields that need more robust, processable, and multifunctional materials. However, the full potential for the use of CNTs as composite reinforcement is still hampered by several problems. The major challenges lie in two aspects, (i) homogeneous dispersion of CNTs in the polymeric matrix and (ii) efficient contact between polymer and CNTs on the surface of two components.[10] The key factor to meet above requirements is to improve the interfacial interaction between the polymer and CNTs. When the load transfer between the CNTs and the matrix mainly comes from electrostatic and Van Der Waals interactions,[11] these interactions are very weak. So good interfacial chemical bonding is necessary for improving the physical and chemical properties of CNTs/polymer composites.

Collagen, a biopolymer whose stalks consist of right-handed supercoils of three left-handed polyproline II-type helices with major sequences of (Gly-Pro-Hyp)$n$,[13] is a bioelectret and contains great number of dipoles and molecule-bound charges stored in its molecule. It has the properties of polarization, electrostatic attraction, etc.,[14] and is one of the most abundant and ubiquitous proteins in the human body and a natural biomaterial commonly used in tissue engineering and clinical medicine.[15] However, the use of collagen as a matrix in tissue engineering is limited because untreated collagen is mechanically weak and has very low thermal stability. Therefore, extensive efforts have been made to mimic[16] or stabilize[16–18] its soft conformation. Although these approaches were successful in gaining its mechanical stability, rarely research has focused on improving both the thermal and the mechanical stability with a simple procedure. Here we proposed a novel composite by use of chemical bonding to combine multiwalled carbon nanotubes (MWCNTs) with grafted collagen matrix. The model of spatial interactions existing in this nanocomposite was shown in Figure 1. Since the surface of functionalized MWCNTs contained abundant –COOH and –OH,[19] they could be anchored with ultrasonic vibration into polymer matrix by covalent or hydrogen bond without need of peptide coupling agent.[20] This composite possessed distinct thermal stability and good mechanical properties resulted from the synergistic effect between its two components. The synthesis strategy was facile and would therefore be used as a model system to study the influence of the CNTs to polymer matrices.

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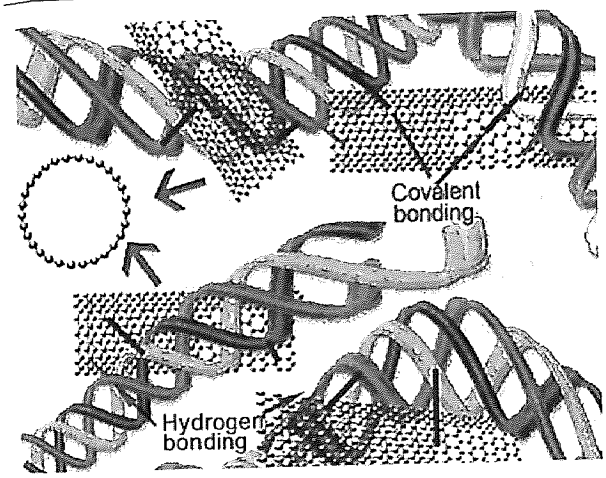


Fig. 1. Schematic of grafted collagen-MWCNTs composite, the carboxyl, and hydroxyl groups existing on the surface of MWCNTs can form covalent and hydrogen bonds with amino groups dangling in collagen chains, respectively.

## 2. EXPERIMENTAL DETAILS

### 2.1. Preparation and Functionalization of the Carbon Nanotubes

High-purity MWCNTs (purity: 92.5–97.1%) obtained through a catalytic chemical vapor deposition followed with a thermal treatment at 2800 °C for 30 min in an argon atmosphere exhibited curly and long tubular morphology (see the SEM images in Fig. 5). The functionalization procedure was as follows: 20 mg as-synthesized CNTs were immersed into 1 M hydrochloric acid for 3 hours and 2.6 M nitric acid for 4 hours, respectively. The obtained products were dispersed in ethanol for further experiments.

### 2.2. Preparation of Grafted Collagen Matrix

The 1:1 mixture of 0.01 mol/l ammonium cerium nitrate (CAN, 99.9 wt%) in 1 M nitric acid and 0.01 mol/l 2,2′-azo-bis-iso-butyronitrile (AIBN, 99.9 wt%) in methanol was used as polymerization initiator. 2.0 g collagen powder with the Gly, Pro, Hyp, and Lys contents of 32.3%, 11.1%, 6.9%, and 4.2% was dissolved in 100 ml water/methanol of 3:1. The calculated amount of methyl methacrylate (MMA) monomer (0.5 mol/l) was added into the solution followed by the calculated amount of initiator. The reaction was carried out at 50 °C in a nitrogen atmosphere for 2 hours. The resultants were then separated by filtration, and the obtained product was washed with distilled water and extracted with acetone to remove the loosely bound homopolymer. This process was continued until no homopolymer was founded.

### 2.3. Preparation of Grafted Collagen-MWCNTs Nanocomposite

Functional MWCNTs and the grafted collagen were mixed at different weight ratios and completely dispersed in ethanol by vigorous stirring for 4 hours. After ultrasonic vibration for 4–6 hours the grafted collagen-MWCNTs nanocomposites were obtained. After filtrated the final products were washed thoroughly with absolute ethanol and evaporated in room temperature to remove impurities and solvents.

### 2.4. Characterization

The FT-IR spectra were recoded on a Nicolet 5ZDX spectrometer. The tensile tests were carried out on an Instron universal material testing system (model 5567) at room temperature. Scannning electron micrographs (SEM) images were obtained with a Hitachi X650 operating at an accelerating voltage of 20 KV. The sample thermal analysis was characterized by using a TGA/DTA apparatus (TMDSC, SDT600, TA instrument) operated at a heating rate of 20 °C/min, to determine simultaneously the reaction heat of materials and the correlation between temperature and weight lose. In these experiments, 5–30 mg samples were placed under helium inside a sealed chamber, and their weight and temperature were monitored closely with thermocouples situated under the sample and the reference, respectively. When the test began, the samples were heated to 100 °C and maintained at this temperature for 20 min to remove the thermal history. The temperature was finally increased to 900 °C. The temperature and the mass of both the samples and the reference were monitored at all the time. Infrared emissivity testing procedure was carried out as follows: phenolic–aldehyde acetal adhesive was used as adhesion agent, dimethyl benzene was used to adjust the density of adhesive with a proportion of 1:2 (v/v), composite powder samples were dispersed uniformly in mixing glue with the volume ratio of 25–30%, the powder entrapped glue was brushed on a square polishing aluminum foil ($60 \times 60$ mm$^2$). After the coating surface was set in air for 1 hour and then was exerted by a pressure of 0.1–0.2 MPa for 2 hours, the infrared emissivity of these coatings was recorded using IR-I infrared emissivity measurement instrument (supplied by Shanghai Research Institute of Technology and Physics, China).

## 3. RESULTS AND DISCUSSION

### 3.1. FT-IR Characterization

To identify the interaction between the MWCNTs and grafted collagen, the FT-IR spectra of three matrices were measured (Fig. 2). The functionalized MWCNTs showed a middle stretching adsorption peak resulting from –COOH at 1640 cm$^{-1}$,[21] while the FT-IR spectrum of the grafted collagen displayed two characteristic infrared absorbance peaks of amide I and amide II of collagen at 1644 and 1559 cm$^{-1}$, respectively. The peak of amide II weakened and the peak of amide I violetshifted for 14 cm$^{-1}$ after the functionalized MWCNTs reacted with grafted collagen. The adsorption peak at 1630 cm$^{-1}$ might be due to the

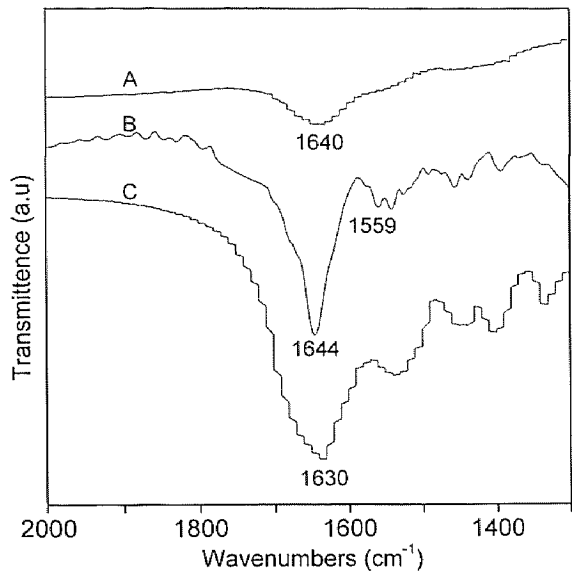


Fig. 2. FTIR spectra of (A) functionalized MWCNTs, (B) grafted collagen, and (C) grafted collagen-functionalized MWCNTs nanocomposite.

stretching vibration of the C=O group of the amide functionality in MWNT-$CONH_2$.[22]

### 3.2. Mechanical Properties Tests

Figure 3 shows the stress–strain curves of the MWCNTs-grafted collagen nanocomposites. The related mechanical properties were summarized in Table I. Representatively, no noticeable yield for both the collagen and grafted collagen was observed (curves a and b) due to high flexibility of these materials. When 2.0% (wt%) MWCNTs were doped into grafted collagen matrix, a clear yield, and postyield period could be observed, which was due to the fact that carbon nanotubes evidently improved the rigidity of grafted collagen. When the collagen peptide chains were grafted the tensile stress increased by about 8.9% (from

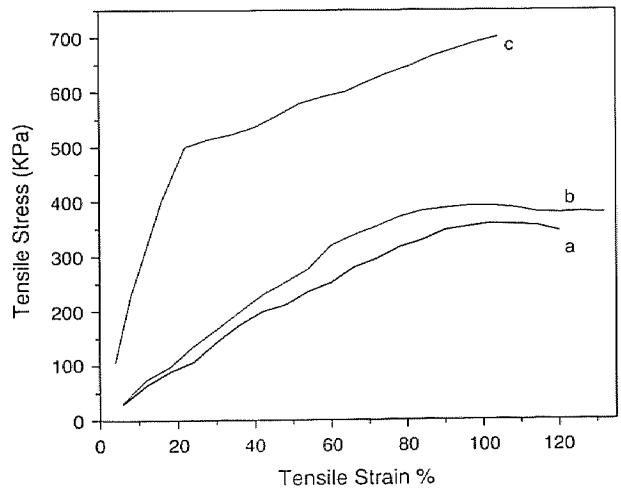


Fig. 3. Tensile stress–strain curves of collagen (a), grafted collagen (b), and grafted collagen-2.0% MWCNTs (w/w) composite (c).

Table I. Mechnical properties of nanocomposites.

| Samples | Maximum tensile strength (KPa) | Yield strength (KPa) | Tensile modulus (KPa) |
|---|---|---|---|
| Collagen | 359 | 14 ± 0.9 | 328 ± 33 |
| Grafted collagen (GC) | 391 | 16 ± 1.1 | 401 ± 29 |
| GC/0.2%MWCNTs | 548 | 21 ± 1.0 | 586 ± 31 |
| GC/0.5%MWCNTs | 567 | 24 ± 0.7 | 647 ± 28 |
| GC/1.0%MWCNTs | 602 | 26 ± 0.7 | 688 ± 36 |
| GC/2.0%MWCNTs | 655 | 27 ± 0.5 | 727 ± 45 |

359 to 391 KPa) in comparison with the bulk collagen, while the elongation at break point could be raised by 5.3%. When the collagen matrix was doped with MWCNTs, the tensile properties were greatly improved. The tensile stress increased gradually when more nanotubes were doped. For samples, the tensile stress at a MWCNTs content of 2.0% w/w showed an increase of about 95.2% (from 359 versus 701 KPa). Unfortunately the elongation at break point decreased gradually, which indicated the composite became more fragile. The acceptable reason was that the MWCNTs reduced the plasticity of polymer matrix. When more MWCNTs (more than 2.0%) were incorporated into the matrix, the tensile stress increased very slightly, this was probably due to the limited bonding of the matrix with MWCNTs.

Figure 4 shows the curves of yield strength and tensile modulus vs MWCNTs content (wt%). The doping of 2.0% w/w MWCNTs resulted in an increase of the yield strength of collagen by 92.9% (from 14 to 27 KPa) and an improvement of the elastic modulus by about 92.3% (from 328 to 727 KPa), respectively. This result was similar to the previous studies on clay enhanced polymer nanocomposites.[23, 24]

### 3.3. SEM Micrograph

The MWCNTs was well dispersed with diameter range from 50 to 100 nm (Fig. 5). The SEM image of single

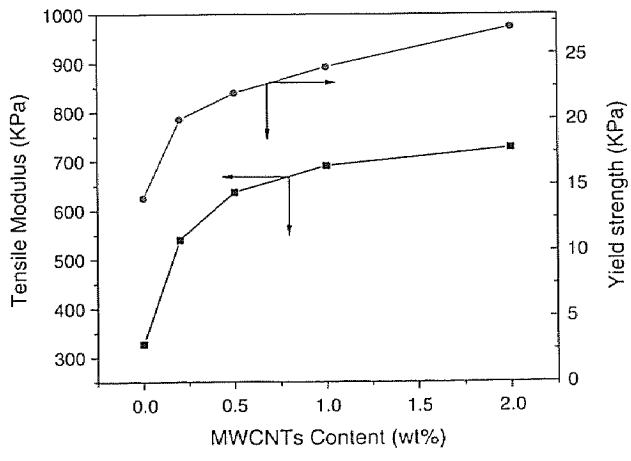


Fig. 4. Tensile modulus (•) and yield strength (■) versus MWCNTs (wt%).

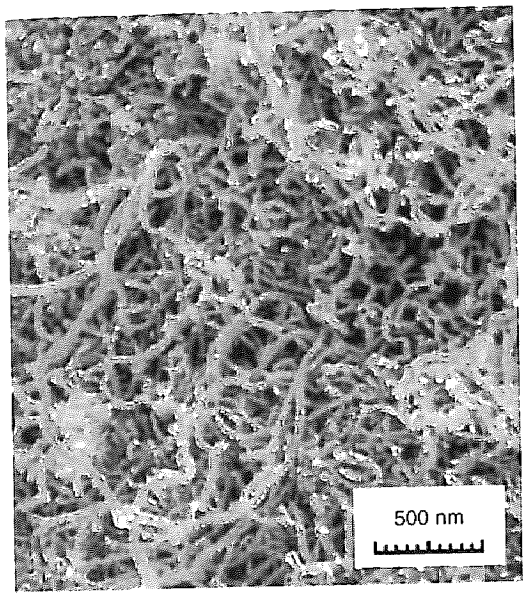


Fig. 5. SEM micrograph of multiwalled carbon nanotubes.

functional MWCNT indicated it contained defects and curvatures on the side wall (Fig. 6(A), indicated with arrow). These defects and curvatures could provide useful sites for chemical or physical functionalization of CNTs.[25] The SEM image showed clearly that a homogeneous dispersion of MWCNTs was achieved throughout the grafted collagen matrix. Most of MWCNTs were touched or wrapped by

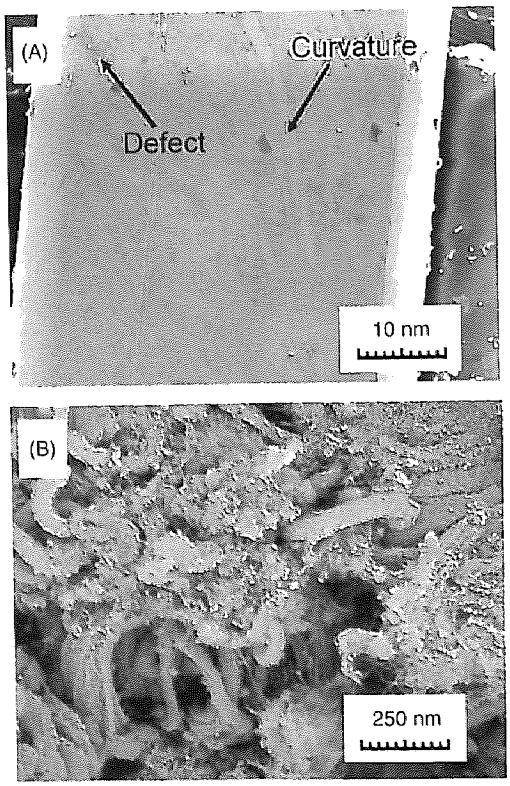


Fig. 6. SEM of (A) single functional MWCNT and (B) micrographs of grafted collagen-MWCNTs composite.

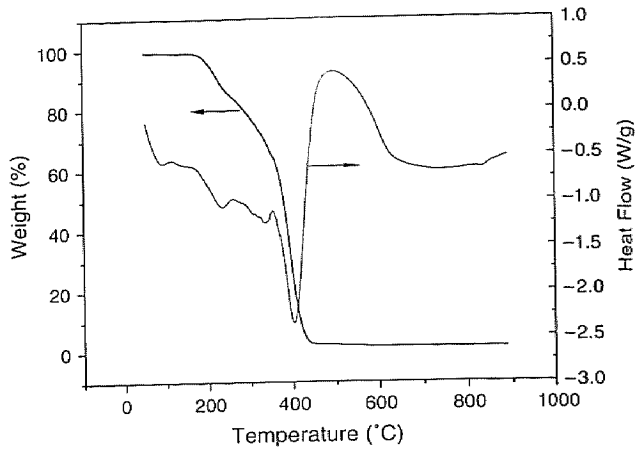


Fig. 7. TGA/DTA graphs of grafted collagen-2.0% MWCNTs composite.

grafted polymer chains and the collagen chains could attach on the sidewall of MWCNTs.[26] This morphology might give another proof to verify the strong interaction between two components in microscale.

### 3.4. Thermal Analysis

The changes of thermal properties during heating of samples at temperatures ranging from room temperature to 900 °C in air at a rate of 20 °C/min could be examined by thermal analysis. The TGA patterns of typical grafted collagen-2.0% MWCNTs composites were shown in Figure 7. Thermal weight loss of the composite was gradual in the temperature range from 178 to 430 °C. As known, the original degradation temperature of the collagen is 39 °C only,[27] thus the grafted collagen-MWCNTs composites possessed better thermal stability than collagen. The DTA curve of grafted collagen-2.0% MWCNTs presented some tiny endothermic peaks before 350 °C, which were due to the destruction of complicated link state between grafted collagen matrix and MWCNTs. A strong endothermic peak occurred at 395 °C, which was associated with the decomposition of covalent bonding between the biopolymers and MWCNTs.[28] So the DTA behavior of the composite gave a proof to verify strong interfacial interactions between two components. The single exothermic peaks at 494 °C were attributed to the pyrolysis of the collagen biopolymer.[29]

### 3.5. Infrared Emissivity Testing

The data obtained from infrared emissvity testing were shown in Table II. It was observed that the infrared emissivity (8~14 $\mu$m) of nanocomposites was much lower than those of two components (grafted collagen-2.0% MWCNTs: 0.570, grafted collagen: 0.896, MWCNTs: 0.798). The acceptable reason was also assigned to the strong synergistic effects between grafted collagen and MWCNTs. These effects changed the transfer modes o

**Table II.** Infrared emissivity (IE, 8~14 $\mu$m) of products.

| Samples | GC | MWCNTs | GC-MWCNTs (0.2%) | GC-MWCNTs (0.5%) | GC-MWCNTs (1.0%) | GC-MWCNTs (2.0%) |
|---|---|---|---|---|---|---|
| IE | 0.896 | 0.798 | 0.598 | 0.587 | 0.582 | 0.570 |

dangling bonds lain in the sidewall of CNTs, which made the absorption bands existing in 8~14 $\mu$m weaken.[30] Thus the whole material possessed lower emissivity than that of two components alone.

## 4. CONCLUSIONS

A novel nanocomposite is prepared by covalently attaching MWCNTs on a grafted collagen matrix. The obtained MWCNTs displayed well-distributed morphology with the diameters ranging from 50 to 100 nm. Based on a supersonic doping process, MWCNTs are homogeneously dispersed in the grafted collagen matrix. The incorporation of 2.0% (w/w) MWCNTs in the matrix results in an increase of approximately 95.2% in the tensile stress, an increase of yield strength of the collagen by 92.9% and an improvement of elastic modulus by about 92.3%, respectively. MWCNTs can also improve the thermal stability of collagen. Besides, the nanocompsites possess much lower infrared emissivity value than that of its two components, which is attributed to the synergistic interactions between MWCNTs and collagen matrix.

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