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Strategies for transmission electron microscopy specimen preparation of polymer composite

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FOREWORD

This special publication is one in a series stemming from the National Nanotechnology Initiative (NNI) Nano-EHS Research Strategy, which identified Nanomaterial Measurement Infrastructure as one of the essential areas of research needed in order to develop an effective risk assessment and management plan regarding various aspects of nanotechnology in consumer products as it pertains to human health, exposure and the environment. The National Institute of Standards and Technology (NIST) was identified as a lead agency in the development of measurement strategies for the detection and characterization of nanomaterials within commercial products. Carbon nanotubes in commercial products, the focus for this special publication, were one of the nanomaterials identified in the Nano-EHS research strategy.

The current guideline presents a strategy for preparing thin section specimens of multiwall carbon nanotube polymer composites for transmission electron microscopy and scanning transmission electron microscopy. Model system explored is a multiwalled carbon nanotube epoxy composite but the strategy should be generally applicable to other types of polymer nanocomposite materials. Updates to this protocol may be released in the future. Visit http://www.nist.gov/mml/nanoehs-protocols.cfm to check for revisions of this protocol, or new protocols in the series. We also encourage users to report citations to published work in which this protocol has been applied.

1. Introduction

Carbon nanotubes (CNTs) are commonly incorporated into polymers to improve mechanical, electrical or thermal properties of the composite material. Growing commercial and industrial interest in CNTs is well reflected in the increased global CNT production capacity and the rapidly increasing number of CNT-containing consumer products [1-3]. As the CNT market expands and the number of commercial applications for CNTs grows, efforts to detect, characterize and analyze nanocomposite materials during their commercial lifecycle is increasing. Detection and characterization of CNTs within commercial products, however, can be quite challenging. CNT type, surface functionalization, and concentration; formulation used for the polymer matrix material; and the identity of other additives which are usually deemed proprietary by the manufacturer can influence the utility and effectiveness of analytical methods used to study these materials.

To facilitate characterization and analysis of CNTs in nanocomposite materials, the National Institute of Standards and Technology (NIST) has developed a series of guidelines describing basic procedures for material fabrication, specimen preparation, and nanomaterial characterization using several commonly applied techniques and methods. The procedures presented in this series focus on a simple nanocomposite system of multi-walled CNTs (MWCNTs) and epoxy. While there are many different types of CNTs and polymers used to produce nanocomposite materials for a variety of commercial applications, the epoxy based MWCNT nanocomposite is one of the most widely used nanocomposite system and the preparation and analytical challenges associated with this system is generally applicable to other polymer nanocomposite systems.

2. Principles and Scope

Transmission electron microscopy (TEM) is the main technique for nanomaterial characterization, due to its ability to provide morphological and chemical details of materials at sub-nanometer resolution. Scanning electron microscopy (SEM) analysis of nanocomposite materials is useful and widely used because it is one of the few high resolution imaging techniques that can differentiate carbon nanotubes from other similar structures or particles with similar aerodynamic sizes. However, TEM is needed to analyze individual nanotube structures such as number of walls, diameter of hole, etc., and to characterize surface coating or functionalization of tubes. The purpose of this special publication is to provide a detailed procedure for fabricating thin nanocomposite sections for TEM analysis using *in situ* and *ex situ* focused ion beam (FIB) lift out techniques. Detailed discussion of TEM, SEM and FIB theory and operation is beyond the scope of this publication and can be found in the following references [4-6].

3. Terminology

Carbon nanotubes – Seamless cylinders of one or more layers (single-wall, SWCNT, or multi-wall, MWCNT) of graphene with open or closed ends.

Nanocomposites – Composite material formed by incorporating nanomaterial as filler into a (polymer) matrix.

Focused Ion Beam Scanning Electron Microscope (FIB SEM) – Dual platform instrument equipped with a focused ion beam column and an electron column. For the Transmission Electron Microscope (TEM) specimen preparation process, FIB is mainly used for material processing and SEM is used for imaging.

Slotted half grid – A copper half grid specially designed for an *ex situ* lift out process. This type of grids allows additional thinning and FIB processing of *ex situ* lift out TEM specimens.

TEM thin section – A lamella of material that is sufficiently thin (100 nm or less) to be electron transparent. Typical TEM thin sections are <10 μm wide and <10 μm tall.

TEM half grid – A roughly semicircular (≈3 mm in diameter) piece of thin metal substrate used for mounting TEM thin sections. Copper half grids are the most common type but other types of half grids are also available.

Lift out technique – A method where a rectangular or triangular prism-shaped volume of material (roughly 1 μm thick by 10 μm wide by 10 μm tall) is isolated, lifted out of the bulk material, and mounted on a TEM half grid or slotted half grid.

Micromanipulator – A < 10 μm diameter needle-shaped probe attached to motors with micrometer or sub-micrometer control used for lift out process. This is a common FIB SEM component.

Gas Injection System (GIS) – FIB SEM component used for site specific material deposition or removal.

SEM sample stub or SEM pin stub – Mounting hardware used to hold samples to be analyzed/prepared in a FIB SEM. The shape and size of a stub varies depending on the make and model of the FIB SEM instrument used but they are usually made of aluminum.

4. Materials, equipment and instrumentation

4.1 Materials

4.1.1 Epoxy/MWCNT composite sample
4.1.2 Double sided conductive tape or conductive paint
4.1.3 Aluminum sample stub for mounting the nanocomposite sample
4.1.4 TEM half grids

4.2 Sample Preparation equipment

4.2.1 Stereo microscope for mounting the nanocomposite sample onto the sample stub and TEM half grid onto a grid holder
4.2.2 Sputter coater
4.2.3 Tweezers

4.2.4 Razor blade
4.2.5 Latex or nitrile gloves

4.3 Instrumentation

4.3.1 Focused ion beam scanning electron microscope equipped with a gas injection system and a micromanipulator. The micromanipulator can either be mounted within the FIB SEM chamber for *in situ* lift out or be a standalone micromanipulator system for *ex situ* lift out.

5. Specimen Preparation for TEM analysis of CNT composites

5.1 Sample Mounting

5.1.1 Sample size and geometry

The concentration of CNTs in commercial nanocomposites is relatively low (< 5 % mass fraction of CNTs) and many CNT nanocomposites are poor electrical conductors unless they are specifically engineered as a conductive material. To prevent excessive charging during the TEM specimen preparation process, it is recommended that the sample size be minimized (≈5 mm or less in lateral dimension). A small piece of material can be readily cut from the bulk material using a razor blade. Additionally, having a relatively flat and planar geometry (*e.g.* small piece of nanocomposite film) will facilitate an easier specimen preparation process.

5.1.2 Sample mounting

Sample mounting hardware/stub varies depending on the type of FIB SEM used. The small piece of nanocomposite can either be attached to the stub using a double sided conductive tape (*e.g.* carbon tape or copper tape) or using a conductive paint such as carbon paint or silver paint. If using a conductive paint, use a small amount of paint to prevent the paint from wicking over the sample.

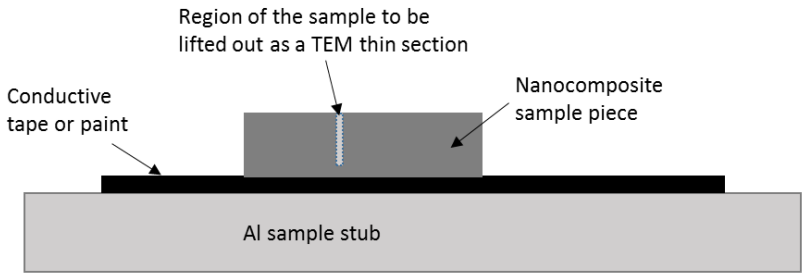

Figure 1: Schematic showing sample mounting geometry.

5.1.3 Sample coating

Coat the mounted nanocomposite sample with a thin layer of conductive material such as Au, Au/Pd, Pt, etc. using a recommended sputter coating procedure. If the top ≈100 nm of the nanocomposites sample is of interest, then a continuous sputter coating is recommended to avoid FIB ion implantation damage during the FIB process [7].

5.2 FIB *in situ* lift out

Basic FIB *in situ* lift out steps for TEM specimen preparation are well established and clearly described in the following patents and articles [8-12]. The main challenge in preparing a TEM specimen of nanocomposite material is making a thin section of nanocomposite material without causing significant damage in the prepared specimen [13]. The following sections describe the general procedure for preparing a TEM specimen from a polymer nanocomposite material.

It is important to use low electron beam energies (≤ 5 keV) and electron beam currents (≤ 200 pA) for the process. An electron beam energy of ≤ 2 keV and an electron beam current of ≈100 pA are recommended, if possible. Polymeric materials may be damaged by either the electron or the ion beam and imaging of the samples should be minimized. If the condition of the lift out specimen should be monitored or documented, it is recommended to use the shortest pixel dwell times and the largest pixel size that are minimally acceptable.

The microsampling and the total release methods are the two commonly used methods for preparing TEM specimens. Both methods are used to prepare thick sections (coupons) and thin sections. However, *in situ* thin section preparation of nanocomposite material is not recommended because the thin sections of polymeric materials tend to warp very easily.

5.2.1 Microsampling Method of FIB *in situ* Lift Out

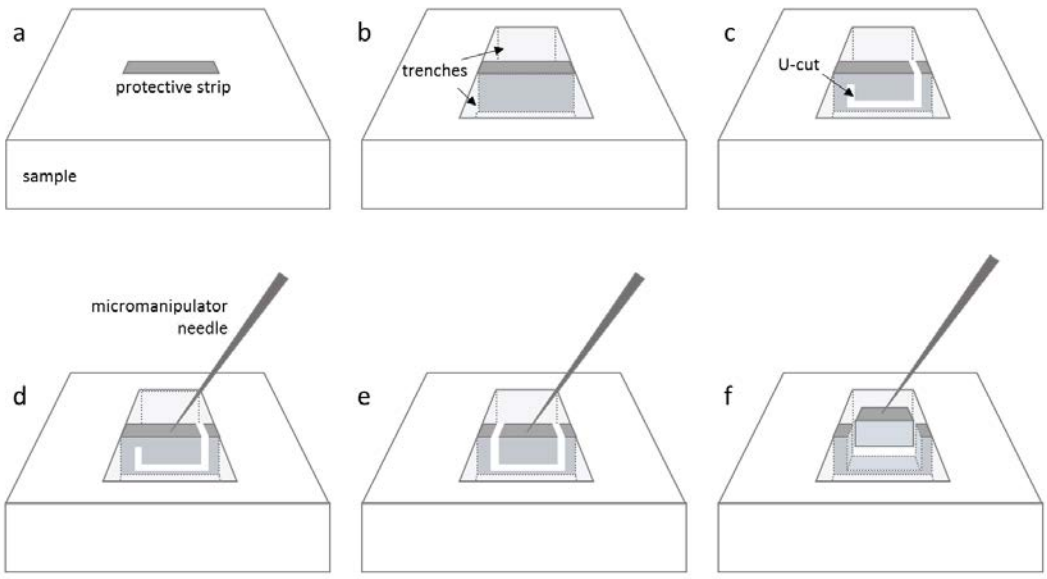

Figure 2: Schematic diagrams of the TEM *in situ* lift out steps adapted from Hitachi microsampling method [8, 11]: a) deposit a protective strip; b) remove the material around the strip, creating a thick coupon; c) partially release the coupon with a FIB milled U-cut; d) attach the micromanipulator needle to the coupon with FIB deposition; e) release the coupon by FIB milling the remaining material away; f) lift out the coupon from the bulk sample.

5.2.1.1 Deposit a protective strip over the region of interest (Figure 2a) using the electron and/or ion beam. Pt, C and W are commonly used as protective coating material. For the purpose of illustration, we will use Pt as the deposition material.

a Identify the region of interest. The width of the TEM specimen should be approximately 10 μm or less because a polymer section wider than this will not be very mechanically stable once it has been thinned to electron transparency.

b If the feature of interest is near the surface of the sample, minimize ion beam damage to the surface by first depositing a thin protective coating layer using the electron beam, for example, a 15 μm x 3 μm x 100 nm (width x height x thickness) strip of Pt layer using 2 keV and ≈3 nA electron beam. In general, use a low beam energy and a high beam current for electron beam deposition.

c Deposit a 12 μm x 3 μm x 1 μm strip of Pt using a 30 kV and ≈0.2 nA ion beam.

5.2.1.2 Remove the material surrounding the region of interest to create a thick coupon (Figure 2b). This process of removing material in the front and back of the coupon is usually called trenching.

a. Use a relatively high ion beam current (several nA and higher) to remove the bulk of the material.

b. Use ≈1 nA ion beam to clean the coupon faces (front and back). If needed, use lower beam currents to clean each face further.

5.2.1.3 Make a U-cut to partially release the coupon from the bulk sample (Figure 2c).

a. This step is normally done with the ion beam at a tilt angle of 45° with respect to the face. Minimize imaging the coupon faces with the ion beam to avoid ion implantation damage.

b. The small tab holding the coupon may not be able to support the weight of the coupon if it is too small. Figure 3 illustrates a) a coupon with an effective U-cut and b) a coupon where the remaining tab is too small. In addition, in example b), additional ion beam processing can unintentionally reattach the partially released coupon to the bulk sample due to redeposition of sputtered material, making the final release and lift out difficult.

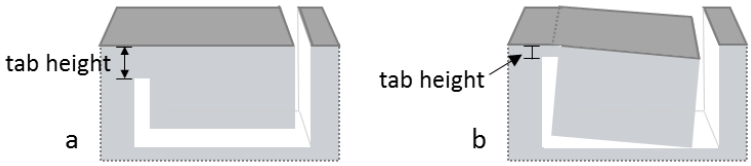

Figure 3: Examples of partially released coupons showing a) sufficient tab and b) insufficient tab to support the weight of the partially released coupon.

5.2.1.4 Attach the micromanipulator needle to the partially released coupon using ion beam deposition (Figure 2d).

5.2.1.5 Release the coupon by FIB milling away the remaining tab (Figure 2e).

5.2.1.6 Lift out the coupon by raising the probe (or lowering the stage) (Figure 2f).

5.2.2 Total Release Method of *in situ* Lift Out

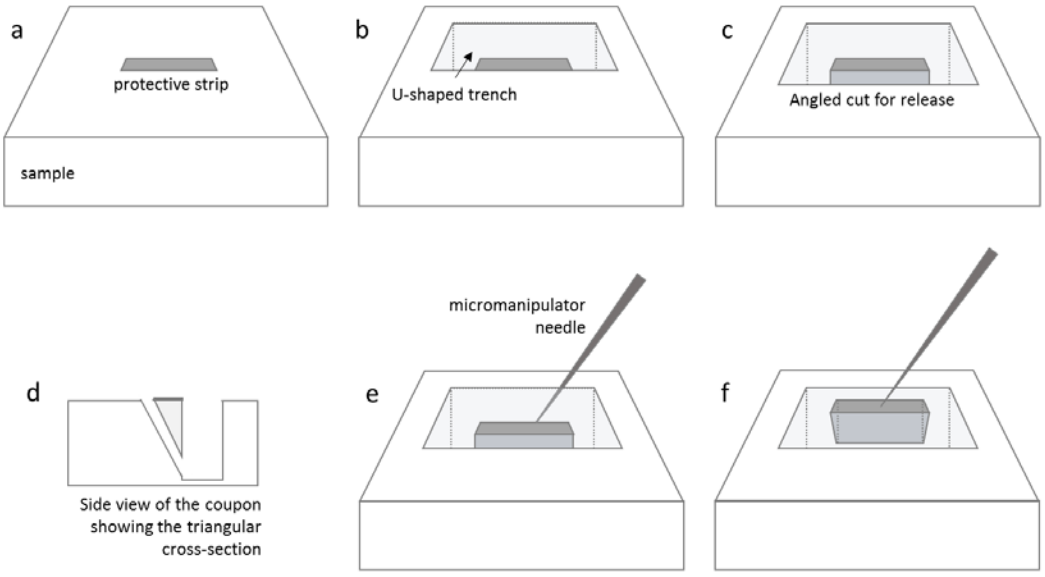

Figure 4: Schematic diagrams of the TEM *in situ* lift out steps adapted from Omniprobe total release method [9]: a) deposit a protective strip; b) remove the material around the strip creating a U-shaped trench; c) release the coupon with an angled undercut, creating a thick triangular prism coupon; d) side view of the sample showing the triangular cross section of the coupon; e) attach the micromanipulator needle to the coupon with FIB deposition; f) lift out the coupon from the bulk sample.

5.2.2.1 Deposit a protective strip over the region of interest (Figure 4a) as described in 5.2.1.1.

5.2.2.2 FIB mill the material surrounding the region of interest to create a U-shaped trench around the region of interest (Figure 4b).

a. Use a relatively high ion beam current (several nA and higher) to remove the bulk of the material.

b. Use ≈1 nA ion beam to clean the coupon faces. If needed, use lower beam currents to polish each face further.

5.2.2.3 Make an angled undercut to completely release the thick triangular prism coupon from the bulk sample (Figure 4c-d). This step is normally done with the ion beam at a tilt angle of 45^{o} with respect to the face. Minimize imaging the coupon faces with the ion beam to avoid ion implantation damage. At this point, the released thick coupon can also be lifted out using an *ex situ* lift out procedure (See Section 5.3.1).

5.2.2.4 Attach the micromanipulator needle to the completely released coupon using ion beam deposition (Figure 4e).

5.2.2.5 Lift out the coupon (Figure 4f).

5.2.3 *in situ* Manipulation to Grid After Microsampling or Total Release Lift Out

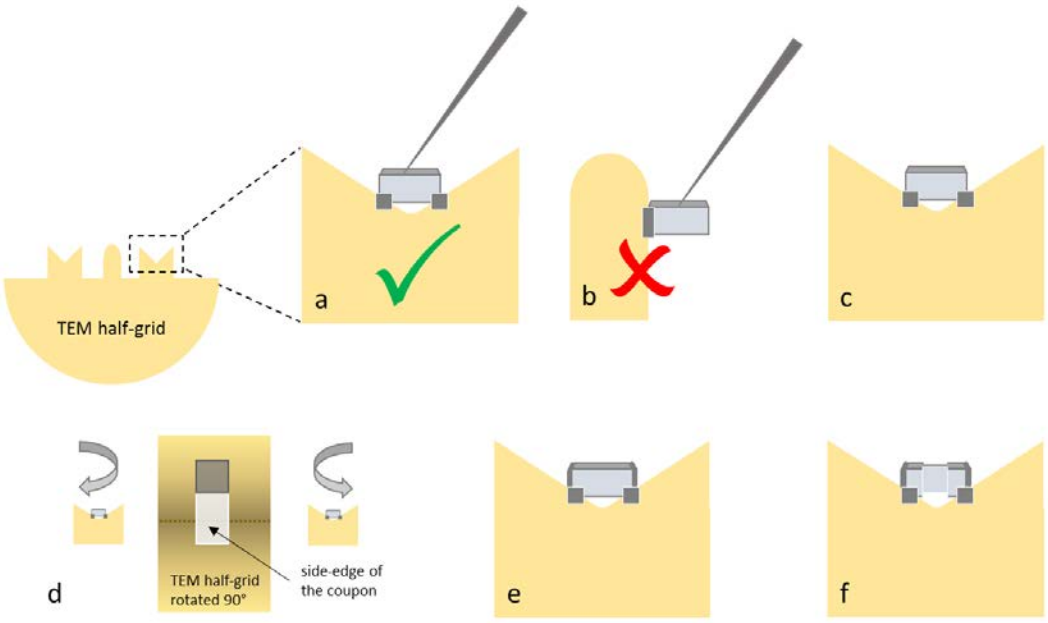

Figure 5: Schematic diagrams of TEM manipulation steps: a) deposit the coupon to a TEM half-grid; c) Mill away the coupon from the micromanipulator; d) rotate the grid to deposit stiffening layers to the side; e) deposit additional protective layer on top; and f) thin and clean each coupon face.

5.2.3.1 Attach the coupon to a TEM half grid using FIB deposition (Figure 5a).

5.2.3.2 Any loose edge of a polymer thin film section will tend to curl and wrinkle. Because of this, the lifted coupon should be supported from the bottom as shown in Figure 4a and the side cantilever mounting (as shown in Figure 5b) of the coupon is not recommended.

5.2.3.3 Mill away the coupon from the micromanipulator (Figure 5c).

5.2.3.4 Ion deposit additional support layers on the side (Figure 5d). This is done with the ion beam at a tilt angle of ≈50° with respect to the face (at 0° stage tilt).

a. Rotate the stage by 90° so the one side of the coupon is visible with the ion beam.
b. Deposit a protective Pt layer on the side.
c. Rotate the stage by 180° so the other side of the coupon is visible with the ion beam.
d. Deposit a protective Pt layer on this side.

5.2.3.5 If needed, ion deposit an additional Pt layer on top of the coupon (Figure 4e). This is sometimes necessary because imaging during trenching and other lift out steps can remove some of the protective Pt layer deposited in step 5.2.1.1.

5.2.4 Final Thinning

5.2.4.1 Thin and clean the coupon with the usual TEM specimen thinning and cleaning procedures (Figure 4f). In general, initial thinning is performed using a 30 kV ion beam with 1 nA or less beam current and the coupon becomes thinner, gradually reduce the beam current to 100 pA or less. A small or negative beam overlap (i.e., large pitch) can help mitigate some of the ion beam induced material damage [14]. With a more beam-robust material, this thinning process could be continued until the coupon is about 100 nm thick before the low kV cleaning steps are performed. However, with a polymer coupon, it is recommended that the 30 kV thinning be stopped when the coupon reaches ≈200 nm in thickness finishing the thinning process using the low kV cleaning steps.

5.2.4.2 Once the coupon is thinned to ≈200 nm, low kV (5 kV and lower) cleaning can be performed to thin the specimen to electron transparency and also remove the damage layer from the high kV ion beam exposure. Refer to Schaffer et al. [12] for detailed thinning parameters for several different examples.

5.2.4.3 If monitoring the thinning process by electron imaging, minimize the beam-induced specimen damage by using low energy (2 keV or less), low current (100 pA or less) electron beam conditions, short pixel dwell times (≈100 ns) and low pixel resolution (1024 x 1024 or lower) imaging parameters.

5.2.4.4 DO NOT (never ever!!!) image the thin section face with an ion beam.

5.3 FIB *ex situ* Lift Out

5.3.1 FIB *ex situ* Lift Out of a Thick Coupon Followed by FIB Thinning

The *ex situ* lift out milling procedures are similar to the FIB *in situ* lift out microsampling procedures. The procedures previously described in Fig. 2a-c are performed. Then each surface is polished to remove redeposition during the U-cut operation *before* milling the tab or material free. This milled-free thick coupon is then lifted out and manipulated onto a slotted half grid using a dedicated *ex situ* lift out station consisting of a zoom light optical microscope and a micromanipulator [14]. Once the coupon is on the slotted half grid, it can be further thinned and cleaned using the steps described in Section 5.2.4. The *ex situ* lift out procedure is well suited for high throughput applications and when an *in situ* micromanipulator is not readily available.

5.3.2 FIB *ex situ* Lift Out of a Thin Section

Although *ex situ* lift out of a thinned section is a commonly used method, thin sections of polymer nanocomposites are, in general, not sufficiently stiff for this method.

6 Abbreviations

CNT	Carbon nanotube
MWCNT	Multi-walled carbon nanotube
TEM	Transmission electron microscopy

SEM	Scanning electron microscopy
FIB	Focused ion beam
GIS	Gas injection system
KeV	Kilo electron volt

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