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Deliverable 6.2

Recipe for TEM sample preparation of organic–inorganic multilayers

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Preparation of cross-sectional TEM lamellae from hybrid organic–inorganic multilayers is highly delicate and challenging. Because, these composites consist of soft organic layers which are deposited on a solid surface with a weak bonding. This low bonding strength and also the soft nature of organic layers make them very sensitive to the TEM sample preparation.

Here different sample preparation techniques were examined to prepare highly durable cross-sectional TEM specimens. We have developed a set of conditions to avoid any potential damage to the sample during the TEM lamella preparation of multilayered composite samples consisting of inorganic zinc oxide (ZnO) layers and organic M13 phage (M13) layers. The conditions in mechanical tripod polishing include choosing (1) The sample surface should be protected with a super glue film in prior to polishing. (2) An appropriate polishing (rotation and oscillation) speed of 50 RPM for the coarse polishing, and then lowering the speed to 20 RPM for fine polishing. After each 50 µm polishing, the sample should be gently cleaned with isopropyl alcohol to remove the debris using a cotton swap. (3) The wedge degree should be set at 1°. (4) Final polishing should continue until a thickness of 20-30 µm is obtained. (5) Isopropyl alcohol should be used as the polishing lubricant instead of water. The use of water must be avoided because water produces high surface tension which increases the shear stress on soft layers. Besides, M13 phages layers can disintegrate in highly polar water. (6) Further thinning is done using an ion beam milling system and an Ar^+ ion gun. To avoid possible damages, samples were thinned with low accelerating voltage (2.6 kV) at an incident angle of 6° from both sides. Also, a liquid nitrogen cooling stage was used to avoid excessive heating of the sample during ion milling.

If these conditions are not met, the M13-ZnO layers might get detached from the substrate (Figure 1a). This was not the case in the method described above, because the protecting layer (glue) on the multilayer system is observed which indicates that the original structure has been preserved during the sample preparation (Figure 1b).

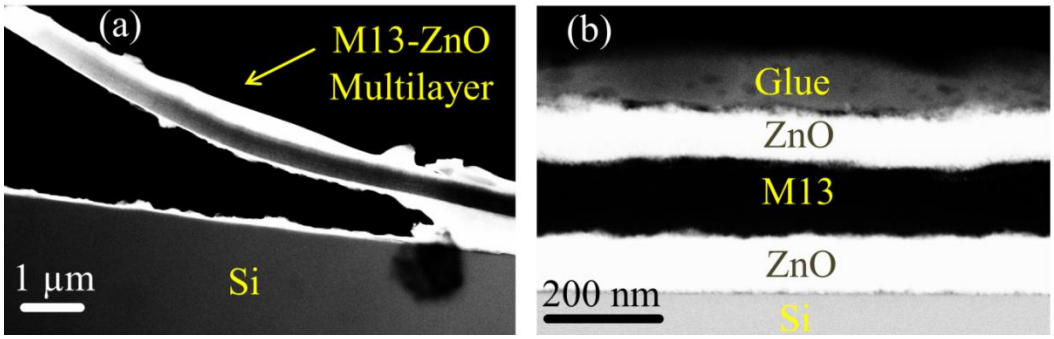

Figure 1. Annular dark-field (ADF)-STEM images of (a) M13-ZnO multilayer structure which is detached from the Si substrate because the conditions of TEM sample preparations were not met. (b) A multilayered lamella prepared by the combined mechanical and ion milling method. The protecting layer (glue) can be observed on the multilayer system.

Figure 2a shows a bright-field (BF)-STEM of a very thin (~ 50 nm thick) bilayered structure consisting of two alternating organic (M13 phage) and inorganic (ZnO) layers on a carbon-sputtered silicon wafer. The sample was protected during the TEM sample preparation using a super glue film. Despite the thinness and sensitivity of the sample, a high-quality sample preparation allowed us to obtain a stable sample for the TEM investigations. A STEM-EDX mapping further confirmed that the two layers of M13 phages and ZnO were formed in an alternating fashion (Figure 2b).

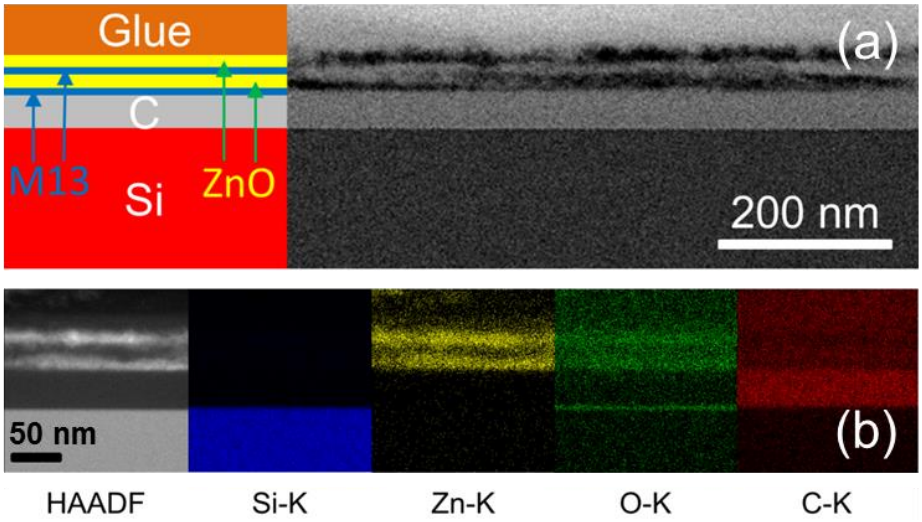

Figure 2. (a) BF-STEM image of the cross-section of the bilayered sample, indicating two thin layers of M13 phages and ZnO (blue and yellow stripes) which were assembled on a carbon-sputtered silicon substrate. An illustration is included showing the layered system. The glue on the top of the specimen was deposited as a protective layer during the TEM sample preparation. (b) STEM-EDX elemental mapping (Si K edge, Zn K edge, O K edge and C K edge) and the corresponding HAADF image of a cross-section of the bilayered sample showing the presence of alternating layers of ZnO and M13 phages on the substrate.