Protocol: Cell Observation (PTA staining)

Required reagents:

1) PBS (phosphate buffered saline)
2) DDW (double distilled water)
3) 1% glutaraldehyde (Nissin EM, Cat. No. 3055) in PBS
4) 2% PTA (Phosphotungstic acid hydrate) (Alfa Aesar, CAS:12067-99-1) in DDW (pH 1.5)
5) Water solution of dextrose (10 mg/ml)

1. 1st day: Culture COS7 cells on ASEM dish.
2. 2nd day: Remove culture medium then wash cells three times with PBS.
3. Fixation: Remove PBS, and incubate in 1% glutaraldehyde / PBS for 30 minutes.
4. Wash three times with DDW.
5. Remove DDW, and stain with 2% PTA in DDW for 30 minutes.
6. Wash three times with DDW.
7. Exchange DDW for dextrose (10 mg/ml) prior to ASEM observation.
8. ASEM conditions: spot size = 30 - 40, acceleration voltage = 20 kV (for example, Fig. 1).

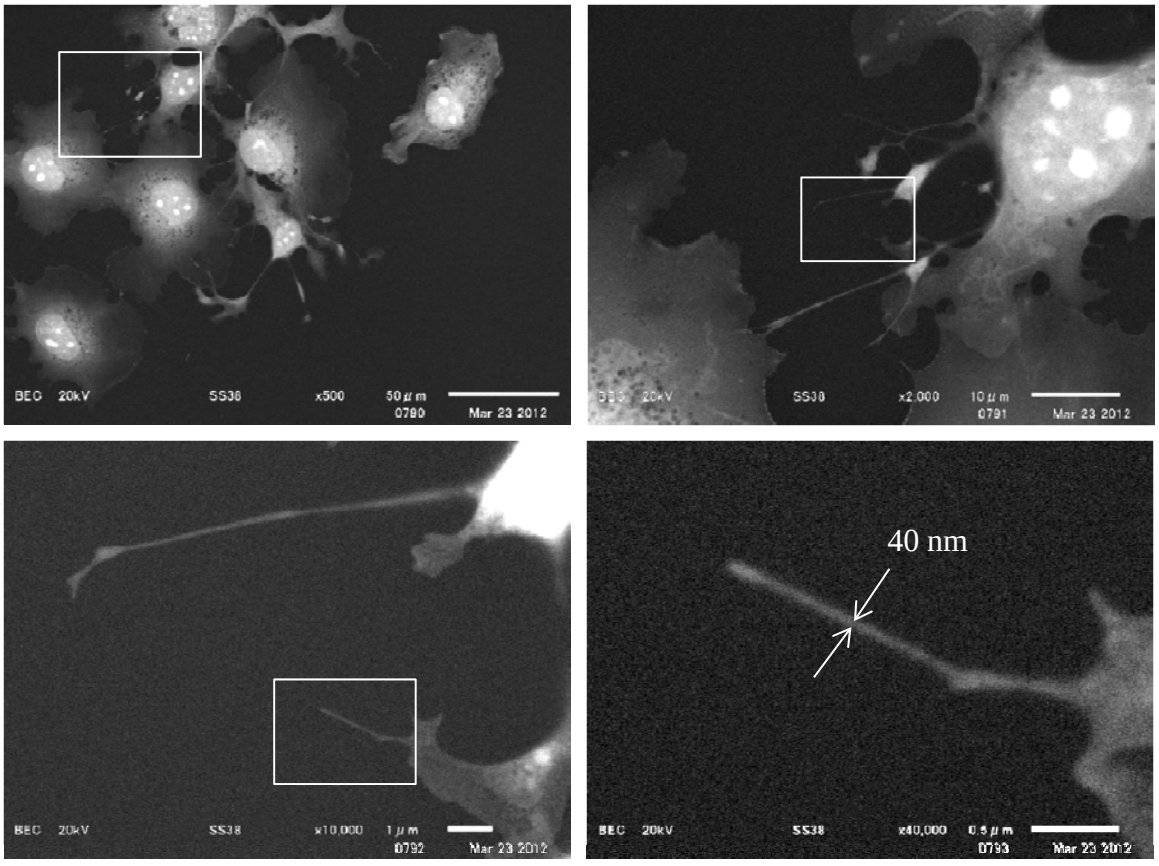

Fig. 1. ASEM images of COS7.

*ASEM : Atmospheric Scanning Electron Microscope